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No. 4 (*Issued December 20th,* 1938).

INVESTIGATIONS OF THE FUNCTIONS OF THE SMALL INTESTINE IN MAN BY INTESTINAL INTUBATION.

PART 1.—THE TECHNIQUE OF INTESTINAL INTUBATION IN MAN.

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IN the past lack of any satisfactory method of approach has greatly hindered the collection of data upon the processes of digestion and absorption in the small intestine of man, and has for the most part limited the work on these subjects to animal experimentation. The recent development by Miller and Abbott of a method of intestinal intubation in man, by means of which an isolated segment of small intestine is rendered available for study, is therefore of great potential value, and it was considered desirable that an attempt should be made to assess its scope and reliability. The method only will be considered here, results obtained being presented in separate papers later, except in so far as it is necessary to mention such findings when discussing the method.

The method of intubating the small intestine used throughout the present work is essentially that developed and described by Miller and Abbott. They used first a tube with two lumina (4 and 5). Later they obtained a suitably flexible tube with three lumina (1), this permitting the development of the method for complete isolation of a segment of intestine. The final technique used in the present work, though similar to the above, differed in certain important respects, especially in that it was found possible to inflate both balloons via the same lumen of the tube, thus leaving an additional lumen which could be used for other purposes, such as the prevention of any accumulation of fluid in the intestine above the isolated segment, or for the instillation at that level of test substances. In view of the possible importance of such differences in comparing experimental

results it is considered advisable to include here a further account of the actual technique used.

Apparatus.

The rubber tube, obtained from the U.S. Rubber Products Company of New York City, was circular in section, and had an outer diameter of 6 mm. and a length of from 2 to 3 metres. The outer wall was approximately 1 mm. in thickness, and the interior of the tube was divided by three radial septa of about the same thickness. The apparatus as used is shown in Fig. 1.

Fig. 1. Diagram showing arrangement of intestinal tube used.

A.—Lumen connected with isolated segment.
B.—Lumen connected with balloons.
C.—Lumen connected with intestine above segment.

Balloons were so attached, one terminally, one more proximally, that on their inflation a segment of small intestine approximately 10 cm. in length was isolated between them. Both were inflated through the same lumen of the tube. Of the two remaining lumina, one was made to open by two or three orifices just above the proximal balloon, the other by numerous orifices into the segment isolated between the balloons. At, and distal to, the upper point of attachment of the proximal balloon only two of the lumina of the tube were needed, and for the attachment of the distal balloon only one lumen. By cutting away the unused sections the flexibility of the terminal portion of the tube could be much increased, thus greatly facilitating the passage of the tube.

Fig. 2 shows diagramatically the apparatus used in maintaining the air pressure in the balloons, and in aspirating samples of juice. The balloons were inflated to constant pressure (35 to 40 cm. of water) with air, by means of the difference in the water levels in the reservoirs A, A'. Continuous aspiration was applied both above and between the balloons by means of a water syphon system, B, B', exerting a negative pressure of about 120 cm. of water. Aspirated fluids were collected in small graduated tubes C, C', and with continuous aspiration, under constant conditions, a moderately steady flow of juice was obtained. A mercury manometer D, and a trap E (to prevent chance cross-contamination between the tubes C, C') were included. Fig. 3 is a photograph of an X-ray film, exposed with the tube in position in the upper jejunum, and with the balloons inflated so as to isolate the 10 cm. segment of intestine.

Intubation.

Co-operation by the subject was necessary, and training improved performance. No difficulty was experienced in the actual swallowing of the tube, though on account of its greater stiffness, and of the presence of the deflated balloons, it was slightly more unpleasant to swallow than a duodenal tube. Once in the stomach no further discomfort was usually felt, and the subjects occasionally went to sleep during the course of the investigations. Straining, retching or coughing might partially empty the balloons, and allow leakage into or out of the segment.
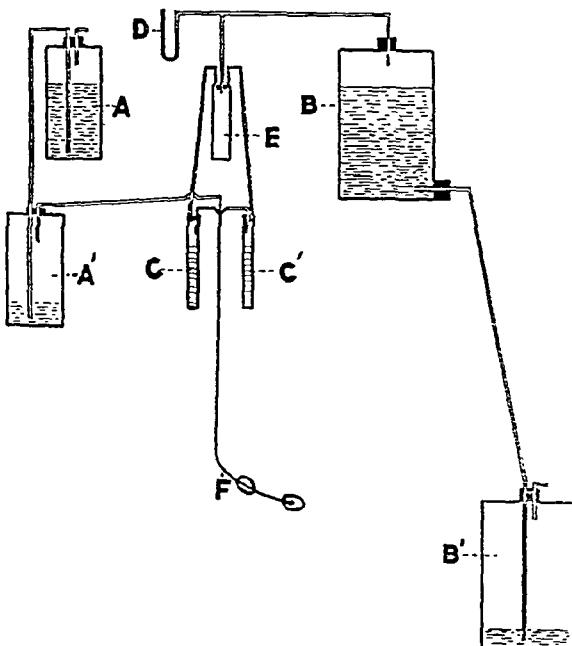
Fig. 2. Diagram showing the apparatus used for the inflation of the balloons, and for the aspiration of intestinal juice from above the isolated segment of intestine, and from the segment itself. A, A′, and B, B′;—air-water reservoirs for the maintenance of constant air pressure in the balloons, and of negative pressure in and above the intestinal segment. C, C′;—graduated · collecting tubes. D ;—mercury manometer. E ;—fluid trap. F ;—the intestinal tube.

The method of passing the tube into the small intestine with the aid of fluoroscopy was similar to that originally described by Miller and Abbott for the tube with two lumina, although the tube with three lumina is definitely less flexible than the former. It was often of advantage to inflate the balloons with about 20 c.c. of air after the tube had been swallowed into the stomach, to facilitate the passage of the terminal balloon to and through the pylorus. After this had been accomplished the balloons were deflated again until the second balloon had passed the pylorus. 20 c.c. of air were then reintroduced

until the desired depth had been attained, when full inflation was obtained by connecting the balloons to the pressure system. The final position of the tube was checked by fluoroscopy.

The time required for reaching the small intestine varied widely. In some cases, especially in trained subjects, the tube would enter the duodenum within 5 minutes ; in others 30 minutes might be required. Occasionally, even in subjects ordinarily presenting no difficulty, the tube could not be induced to enter the duodenum, presumably on account of pylorospasm. Once in the duodenum the tube could be passed on rapidly for another 100 cm., after which its progress slowed down considerably. Usually the tube was down to 150 cm. from the teeth within an hour after it was swallowed. Apparently with sufficient time it could always have been passed to 200 cm. below the teeth, or beyond. This, however, was not attempted in most cases. The greatest depth attained in a normal individual was 220 cm. below the teeth, in about $1\frac{3}{4}$ hours. According to Einhorn (2) the pylorus may be taken as 55 cm., the ileocolic sphincter as approximately 325 cm., below the teeth. With 100 cm. of the tube swallowed beyond the teeth the isolated segment was usually found to lie in the first jejunal loop below the duodenojejunal flexure.

In the first and second parts of the duodenum the motor activity of the intestine was such as to render the method unreliable for most purposes. The intestine at that level also was more sensitive to pressure, and colic readily developed. Below the duodenojejunal flexure the method in general was far more satisfactory, and no trouble arose out of creeping of the tube after full inflation of the balloons, or out of excessive leakage into or from the isolated segment. A useful precaution was to arrange that, as verified by fluoroscopy, the segment isolated lay in an ascending loop of intestine, so that there was less chance of fluid suddenly collecting just above the proximal balloon. By means of the lumen leading to openings above the balloons it was possible to keep the intestine above the isolated segment empty, and to avoid any collection there of fluid which might lead to nausea and vomiting, or to leakage.

In general the balloons could be kept inflated, and observations maintained, for 5 to 7 hours. If, however, much manipulation, or frequent injection of solutions into the isolated segment was necessary the observations could not be continued for as long. Again in one rather highly-strung youth severe nausea, retching or colic almost invariably occurred after $1\frac{1}{2}$ to 2 hours, even with a low air pressure in the balloons (32 cm. of water). Similarly, in an older man with achlorhydria who had had acute alcoholic gastritis in the past with subsequent recurrent attacks of enteritis, nausea very readily occurred, and during one intubation he developed watery diarrhœa. This indicates a definite limitation of the method in the form used, if applied to the investigation of pathological conditions. Irritation of the small intestine, mechanically or chemically, led to a considerable

output of mucus, which might vitiate observations by blocking the tube or by adsorbing substances inserted with consequent difficulty in recovery.

Control of leakage.

When continuous aspiration is applied to the isolated segment the risk of loss due to leakage past the balloons and out of the segment may be taken as negligible. On the other hand there is the distinct possibility of leakage into the segment from above. In spite of all care, leakage sometimes occurred if large volumes of fluid descended from higher levels. The appearance of bile pigments in the fluid aspirated from the segment indicates leakage into the segment from above. In observations involving the oral administration of a readily detectable substance, such as glucose, or its insertion directly into the intestine above the isolated segment, its absence in the material aspirated from the segment may be taken to prove lack of contamination. At other times a similar control may be obtained by inserting above the segment a 0·25% solution of vital red in physiological saline. Even if at once reaspirated, enough vital red solution is left to keep the contents of the intestine above the proximal balloon coloured for a considerable time, and thus leakage into the isolated segment can be detected by the appearance of dye in the segment juice. A further and even more sensitive test for leakage into the isolated segment is provided by determinations (by a modified Wöhlgemuth technique) of diastase. The mixed juice from above the isolated segment always gives high values for diastase, uncontaminated juice from the segment gives relatively low values, usually 1/500 to 1/2000 of the former. Assuming the diastatic activity of the juice secreted by the isolated segment to be zero, a maximal value for any contamination can be calculated. Thus in one typical experiment the mixed juice from above the segment had a diastatic activity of 400 units per 1 c.c., the segment juice an activity of less than 0·7 unit per 1 c.c., and a volume of about 10 c.c. in 15 minutes. Therefore the maximal leakage into the segment must have been 0·018 c.c. in 15 minutes. It was often possible to insert solutions above the isolated segment for periods of 15 minutes or more without any leakage occurring.

Although leakage into the isolated segment from above is thus capable of accurate control, leakage out of the segment following insertion of solutions into it has been difficult to disprove. As mentioned above, it is of advantage to have the final position of the isolated segment in an ascending intestinal loop. Any leakage is then more likely to occur past the proximal balloon, with the possibility of detecting substances initially inserted into the isolated segment in the fluid aspirated from above the segment. Excluding experiments in which colic developed and the increased motor activity of the intestine rendered leakage more likely, the consistency of the results obtained for the absorption of glucose and other substances from the isolated intestinal segment gives evidence of the general reliability of the method. Similar evidence is afforded by the recoveries after short periods of time of substances

less readily absorbed. Thus the recoveries of solutions of vital red, magnesium sulphate, and ferric ammonium citrate introduced into the segment are shown in the table. The colorimetric estimation of vital red was not satisfactory. Recoveries of over 100% were obtained probably on account of errors introduced by opalescence of the return fluids, and this precluded the use of vital red as an accurate quantitative control of leakage. The results of rough nephelometric estimations for magnesium sulphate and estimations of iron by the method of Klumpp (3) at least indicate the general reliability of the isolation of the intestinal segment.

TABLE.

The recoveries of various substances introduced into an isolated 10 cm. segment of small intestine.

Subject.	Segment depth in cm.	Substance inserted.	Time of withdrawal.	Recovered.
L.M.	140	10 c.c. vital red solution	15 min.	114%
L.M.	125	20 c.c. vital red solution	35 min.	113%
L.M.	130	20 c.c. vital red solution	30 min.	104%
H.E.O.	120	10 c.c. 20% $MgSO_4$ solution	5 min.	112% to 96%
L.M.	135	20 c.c. 10% $MgSO_4$ solution	7 min.	92%
L.M.	135	20 c.c. 10% $MgSO_4$ solution	7 min.	117%
I.S.E.	140	40 c.c. ferric-ammonium-citrate solution (=236 mgm. iron)	35 min.	230 mgm. iron
I.S.E.	155	40 c.c. ferric-ammonium-citrate solution (=44·3 mgm. iron)	35 min.	39·6 mgm. iron
L.M.	150	40 c.c. ferric-ammonium-citrate solution (=44·3 mgm. iron)	35 min.	40·2 mgm. iron

Washing out the isolated segment, and recovery of inserted substances.

Whereas it is possible with only a few washings with saline to recover substances such as glucose inserted into the isolated segment, this is not the case with those which are markedly adsorbed by, or induce much secretion of, mucus. Enzymes and many other substances are adsorbed by the intestinal mucus. In order to eliminate initially such adsorbed substances practical advantage may be taken of the fact that vital red also is strongly adsorbed. A satisfactory procedure, followed in most cases, was found to be the initial insertion into the segment of from 10 to 20 c.c. of an approximately 0·5% solution of vital red in saline. The segment was then washed out repeatedly until all colour was absent from the return fluid. It was shown that the concentrations of dye and diastase, for example, fell off in an exactly parallel

manner. Usually as much as 500 c.c. of salt solution must be used, and the time required is a serious drawback. The difficulty is accentuated by the existence of a 5 to 7 c.c. dead space in the tube.

Enzyme determinations.

Particular difficulties were encountered in the studies on the secretion of enzymes to be described in subsequent papers. It was found that the enzyme output could readily be increased by mechanical stimulation, and in association with colic and nausea and probable increased motor activity of the intestine. Thus it was necessary to discard any observations during which colic occurred, and to avoid changes in balloon pressure, or mechanical irritation due to repeated running of solutions in and out of the isolated segment or the proximal bowel above the segment. It also became clear that blowing air in and out of the tube in an attempt to obtain juice contained in the dead space of the tube should be avoided, as it definitely stimulated the secretion of enzymes.

SUMMARY AND CONCLUSIONS.

1. A modification of the method of intestinal intubation originally developed by Miller and Abbott has been described which permits complete isolation of a segment of small intestine in man.

2. With trained subjects, and with the precautions outlined the method has proved practicable for investigational purposes.

3. In studying secretion into an empty segment of intestine it is capable of giving accurate results.

4. In following changes in solutions inserted into an intestinal segment it has proved less strictly controllable, but is probably satisfactory.

5. In investigating pathological conditions its utility is limited when there is increased sensitiveness or motility of the intestine.

6. As a general clinical method its utility appears to be limited, on account of its complexity, and of its dependence on the intelligent co-operation of the subject.

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Fig. 3. X-ray film, showing the intestinal tube in place in the upper jejunum, the isolated segment of intestine being 105 cm. below the teeth. The balloons are inflated with 8% KI solution, to render them opaque to X-rays.

INVESTIGATIONS OF THE FUNCTIONS OF THE SMALL INTESTINE IN MAN BY INTESTINAL INTUBATION.

PART 2.—DETERMINATIONS OF DIASTASE, INVERTASE, EREPSIN, LIPASE AND LACTASE IN THE PURE JUICE OF THE SMALL INTESTINE.

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By the method of Thiry, or some modification of this, the presence of diastase, invertase, erepsin, lipase and lactase has been established in the pure juice of the small intestine of dogs, and these enzymes have also been demonstrated in extracts prepared from the mucosa of the small intestine of dogs and other animals (vide Babkin (1) for numerous references). In adult animals, unless milk-fed, lactase might be absent, or present only in lower concentration than in young animals (Weinland (15)). All investigators have been impressed by the low enzymatic activity of the juice.

In man the juice of the small intestine has rarely been obtained unmixed with secretions from other parts of the digestive tract. Only six records of such cases have been found and these are collected in Table I, together with those of two cases in which the secretion of the large intestine was studied. It will be seen that diastase, invertase, erepsin and lipase have been demonstrated, but not lactase. Ibrahim (7) however found lactase in extracts from the mucosa of the fœtus and of still-born babies. In man, as in animals, only a low enzymatic activity was found.

Methods.

The juice normally found in the small intestine is a mixture of the pure intestinal secretion with secretions from any or all of the glands opening into higher portions of the alimentary canal. In order to obtain pure intestinal secretion, and to compare its enzyme content with that of the normal mixed juices it is necessary to isolate a segment of intestine. The

TABLE I.

The findings reported by former investigators concerning the presence or absence in the pure juice of the small intestine of man of invertase, erepsin, lipase and lactase.

	Diastase	Invertase	Erepsin	Lipase	Lactase
Busch (1858) (3)	+	—		—	
Démant (1879) (4)	+	+			
Tubby and Manning (1891) (14)	+	+		+	
Nagano (1902) (10)	+	+		—	—
Hamburger and Hekma (1902) (6)	+		+	—	
Bickel and Wagner (1934) (2)	+				
Esser (1908) (5); for large intestine	+	—		—	
Orbéli and Sawitch (11); for large intestine	+	+	+	+	—

method of intubation used for this purpose was described in the preceding paper (13).

Subjects. The majority of observations were carried out on two trained subjects. Of these one, L.M., aged 58 years, was healthy and knew of no illnesses in the past; the other, I.S.E., aged 60 years, also healthy, had suffered 3 years previously from an attack of enteritis lasting about 2 weeks, with one recurrence in the subsequent year. Less numerous observations were carried out on 3 other subjects :—H.E.O., a healthy youth aged 20 years; E.C., a nervous man aged 25 years, who was somewhat under-nourished and had an old rheumatic carditis, with mitral stenosis, but no alimentary disorder; J.E.M., aged 29 years, considered in more detail elsewhere (Owles (12)), exhibiting the syndrome of intestinal carbohydrate dyspepsia, but apparently physically fit. Scattered observations were made on several other individuals.

Intubation. A segment of small intestine approximately 10 cm. in length was used. The depth of the isolated segment is designated as the distance along the tube from the teeth to the mid-point of the segment. Material aspirated from immediately above the proximal balloon is referred to as mixed juice. The juice initially aspirated from the isolated segment was similar. Aspirated fluid was regarded as a fair sample of pure intestinal juice only after prolonged washing of the segment with physiological saline, or after various other procedures to assure removal of material adsorbed by the intestinal mucosa, and to exclude leakage into the segment.

Quantitative methods. The material aspirated from the intestine was not centrifuged, as control experiments showed, in agreement with former workers, a diminution in enzymatic activity if there had been any appreciable

cloud of mucus present. Phosphate buffer at pH 6·8 was used in the determinations of enzyme activity, as being well within the range normal to the small intestine (Knott (9); Karr and Abbott (8)), though not necessarily the optimum for each enzyme. In each case, except that of diastase, a bacteriostatic was added (sodium benzoate or toluene). In the case of diastase the incubation period was only 30 minutes, and controls showed that bacterial contamination introduced no significant error. The determinations of invertase, lipase and erepsin could not always be done after exactly 24 hours incubation, but in any given series of observations the incubation time for all tubes was always the same. Values for these determinations were calculated assuming a straight-line function for enzyme activity, plotted against time. This is not strictly correct, but does not affect conclusions as to the relative values for any given series of observations. Moreover, the absolute values for different series varied over so wide a range as to render errors due to this assumption insignificant.

(1) Diastase. Twelve tubes, the first 10 containing graded amounts from 0·05 c.c. to 0·50 c.c. of intestinal juice, the last 2 1·0 and 1·5 c.c., respectively, were set up with 0·5 c.c. of buffer and 1·0 c.c. of soluble starch solution (0·1% or 0·2%), and made up to 3 c.c. with water. The achromic point tested with iodine after 30 minutes at 38°C. gives units of diastase, = mg. of starch, or c.c. 0·1% solution, digested by 1 c.c. of juice.

(2) Invertase. Two tubes were set up, each with 10 c.c. of a solution containing 10% sucrose and 0·2% sodium benzoate, and 5 c.c. of buffer solution, and in one 1 c.c. of unboiled intestinal juice, in the other 1 c.c. of boiled juice as control. Results are given as units = mg. reducing sugar as glucose, by Benedict's method, after 24 hours at 38°C..

(3) Erepsin. Two tubes were set up, each with 10 c.c. of 2% " Difco Bactopeptone," 5 c.c. of buffer solution, and 0·5 c.c. of toluene, and in one 1 c.c. of unboiled intestinal juice, in the other 1 c.c. of boiled juice as control. Results are given as units = mg. amine-acid nitrogen, by Sörensen's formol titration method after 24 hours at 38°C., in comparison with control.

(4) Trypsin. As for erepsin, substituting a 0·5% gelatin solution for the Bactopeptone.

(5) Lipase. Two tubes were set up, each with 2·0 c.c. of neutral 50% emulsion of olive oil in gum acacia, 1 c.c. of buffer solution, and 0·5 c.c. of toluene, and in one 1 c.c. of unboiled intestinal juice, in the other 1 c.c. of boiled juice as control. Results are given as units = mg. oleic acid titrated with N/10 NaOH after 24 hours at 38°C., in comparison with control. The instability of the emulsion prevents precise accuracy, but allows detection of grosser changes.

(6) Lactase. Qualitative tests only were performed, mixing in a fermentation tube 1 to 3 c.c. of intestinal juice, 10 c.c. of a 10% lactose solution, 5 c.c. of buffer solution, and 5 c.c. of yeast suspension, a control tube being set up with boiled juice. Gas formation indicates roughly the amount of enzyme, if any, present.

The diastatic activity of the juice of the small intestine.

Juice collected from above the isolated segment, which might contain saliva and pancreatic juice, as well as bile and gastric juice, gave high values for diastase ; in 13 estimations on the subject L.M., 1000 to 3000 units per c.c., average 1700 units per c.c. ; in 12 estimations on I.S.E., 450 to 900 units per c.c., and once 275 units per c.c., average 600 units per c.c. ; in 2 estimations on E.C., 850 and 1000 units per c.c.. These subjects were considered normal. Three estimations for J.E.M. with carbohydrate dyspepsia gave values of 800, 900 and 1500 units per c.c..

Following the inital inflation of the balloons the juice first aspirated from the isolated segment was similar to the above ; in subsequent samples
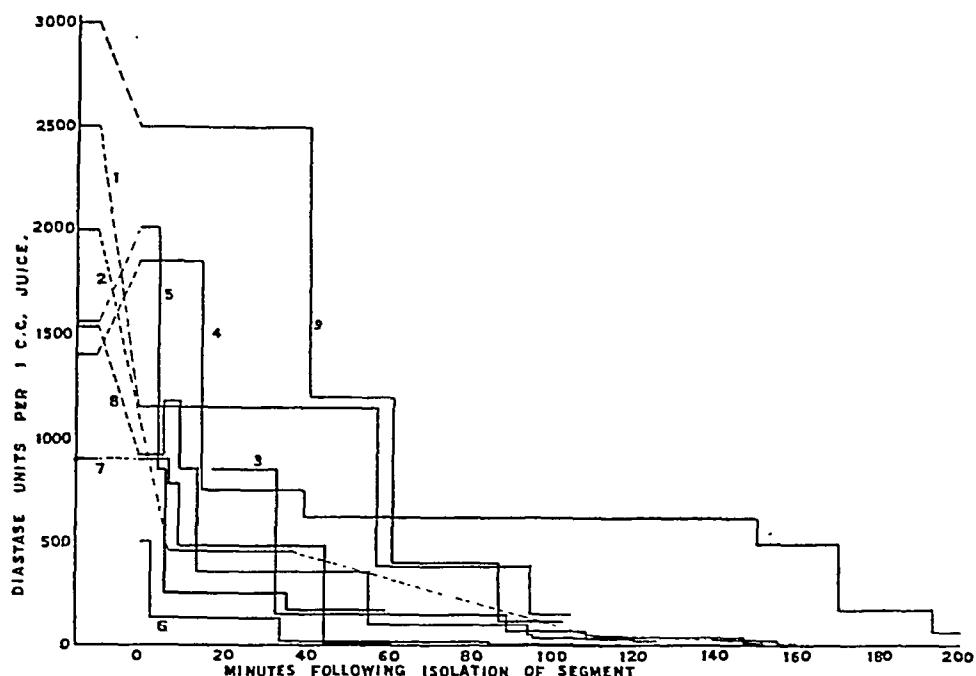
Fig. 1. The diastase concentrations in the juice aspirated after isolating the segment of small intestine. The interrupted lines indicate periods over which no estimations of diastase were made. Observations 1, 2, 3, 4, 5, and 9 upon Subject L.M. ; observation 6 upon Subject I.S.E. : observations 7 and 8 upon Subject J.E.M. (carbohydrate dyspepsia).

the amount of diastase fell progressively to very low figures, usually in 45 to 90 minutes (Fig. 1). Similar results were obtained if total enzyme output per unit time was calculated, taking into account the volume of juice secreted. The fall in diastatic activity was roughly parallel to that of the bile pigment content of the juice, and was accelerated by washing out the segment with saline. If a small volume of a solution of vital red was introduced into the isolated segment, and washing with saline continued until the return fluid was colourless, then after allowing time for subsequent secretion to wash out residual saline from the dead space of the tube, 5 to 7 c.c., later samples of

juice possessed only a very low diastatic activity, 1 or 2 units per c.c., often less, rarely more, and only once or twice as much as 10 units per c.c., compared with initial values of 500 to 3000 units per c.c.. Again it was shown that N/20 hydrochloric acid would in vitro irreversibly destroy all the enzymes studied when added to an equal volume of active juice. If 40 c.c. of N/20 hydrochloric acid were run into the segment then, after removing the acid, subsequent samples of juice again showed low figures for diastatic activity, *e.g.*, 1·5 units per c.c. as compared with 550 units per c.c. before introducing the acid (Table II). Leakage into the segment, as evidenced by the appear-

TABLE II.

The concentrations of diastase, invertase, lipase and erepsin, expressed in units per 1 c.c. of juice, attained in the juice secreted by an isolated segment of small intestine, after initial destruction of enzymes contained in residual mixed juice by the insertion into the segment of N/20 hydrochloric acid.

Subject.	Segment depth in cms.	Diastase			Invertase			Lipase			Erepsin		
		mixed juice A	juice after acid B	A/B	mixed juice A	juice after acid B	A/B	mixed juice A	juice after acid B	A/B	mixed juice A	juice after acid B	A/B
I.S.E.	130	550	1·5	370	0	37	—	194	6	32·5	20	18	1·1
I.S.E.	130	500	1·7	290	59	21	2·8	25	10	2·5	17	18	1·0
L.M.	140	—	—	—	324	23	14·1	54	3	18·0	42	10	4·2

ance in the juice aspirated of vital red which had been inserted into the intestine above the balloons, was invariably associated with higher diastatic activity in the juice. If such leakage occurred, or if a small volume of mixed juice was inserted into the segment, when the diastatic activity of the juice had reached low values, subsequent consecutive samples showed a progressive diminution in diastatic activity from the re-established higher level similar to that from the initial high level (Fig. 2). The fall in diastatic activity with consecutive samples was found to run closely parallel to that of known contaminants, whether occurring naturally (*e.g.*, trypsin), or artifically introduced (*e.g.*, vital red) (Fig. 3), and contrasted strongly with the relative or complete absence of any fall in activity for invertase, erepsin, and lipase.

These facts all point to the cause for the fall in diastatic activity in consecutive samples, and for the low activity in the pure juice obtained by other means, being the removal of some residual contaminant rather than the gradual exhaustion of a store of enzyme from the cells of the mucosa. The possibility that this contaminant was some activator necessary for the diastase of the small intestine was eliminated in 7 control experiments, in which pure juice from the segment was mixed with active mixed juice in definite proportions, the resulting activities corresponding to the amounts present in the ingredients. Salivary diastase is presumably destroyed by
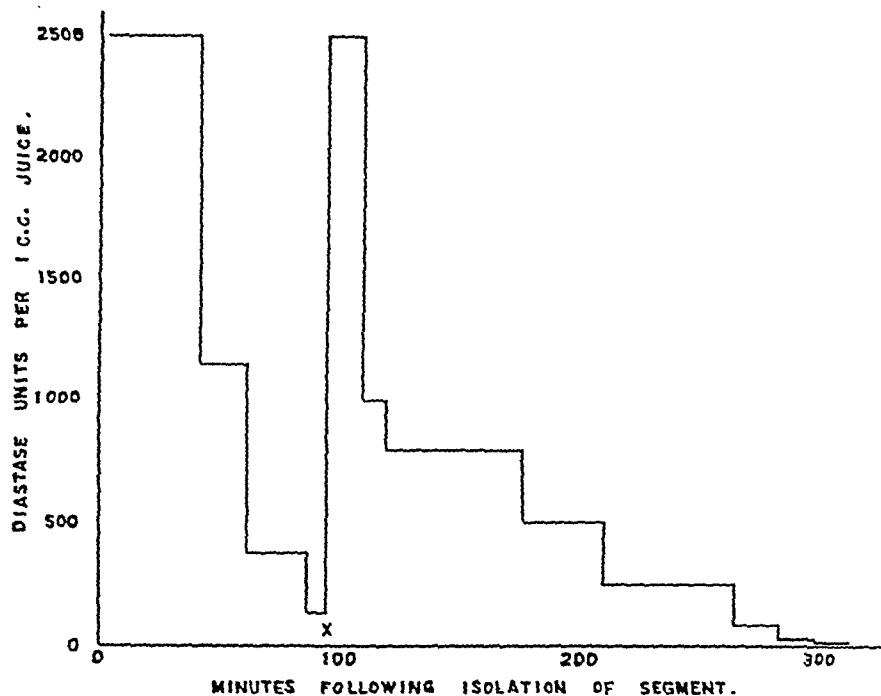
Fig. 2. The diastase concentrations in the juice aspirated after isolating a segment of small intestine. 40 c.c. of mixed intestinal juice were inserted into the isolated segment at the point marked "X." Subject L.M..
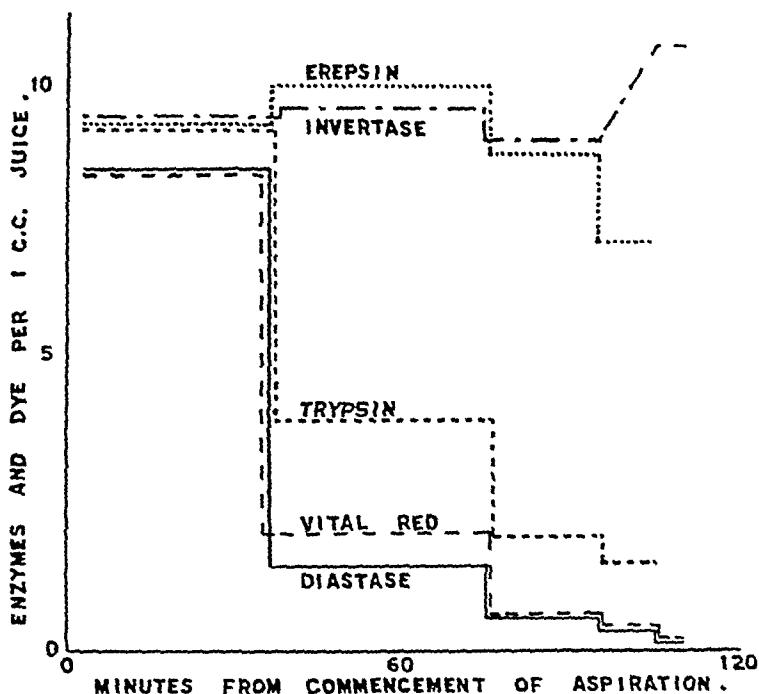
Fig. 3. The concentrations of various enzymes, and of vital red dye, in the juice aspirated after isolating a segment of small intestine, and the initial insertion of a small volume of vital red solution in physiological saline. Subject L.M.. The ordinate scales are *not* in units.

the acid of the gastric juice, which has of itself no diastatic activity, before entering the small intestine, and normal high values were found in the mixed juice in cases in which all saliva was aspirated from the mouth during the observations, and none swallowed. Pancreatic juice is known to possess a high diastatic activity, and is therefore believed to be responsible for the high diastatic activity of the mixed juice. The great difference between the diastatic activities of mixed and pure juice was still found in 4 experiments in which it was tested at pH 7·9, 6·8, 6·6, and 5·0, so that there was no evidence of a second diastase secreted by the small intestine for which the conditions of analysis were unsuitable.

The majority of observations were performed on segments of intestine isolated at depths from the duodeno-jejunal flexure, to 100 cm. below that
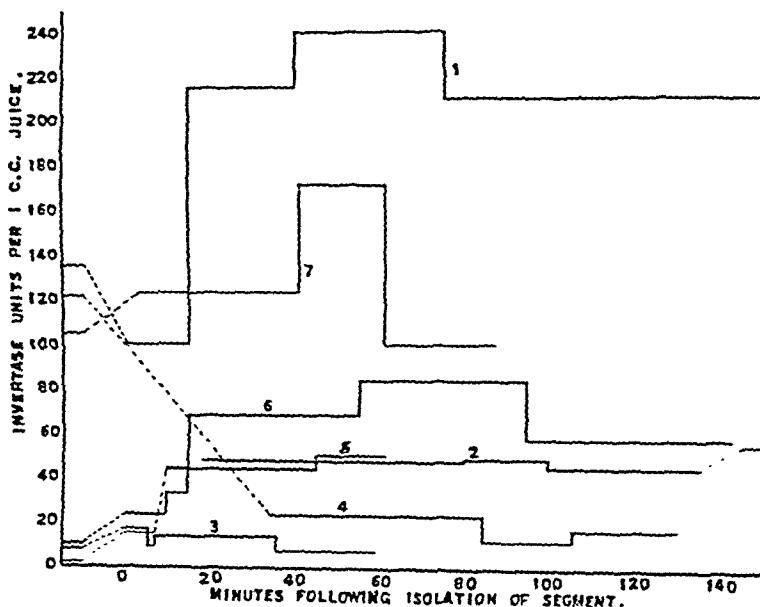
Fig. 4. The invertase concentrations in the juice aspirated after isolating a segment of small intestine. The interrupted lines indicate periods over which no estimations of invertase were made. Observations 1, 2, 3, and 7 upon Subject L.M.; observation 4 upon Subject I.S.E.; observations 5 and 6 upon Subject J.E.M..

level. Over this range no relation was found between intestinal level and the enzyme concentration either of the mixed or of the pure juice.

It is concluded that, in man the diastase of the pure juice of the small intestine is relatively very slight in amount, and can play no significant part in the digestive processes. As it was only about 1/10 of that found in serum (10 to 20 units per c.c.) it is probable that it is not a true secretion of the mucosal cells. The bearing of these facts on the clinical problem of intestinal carbohydrate dyspepsia is considered elsewhere (Owles (12)).

The invertase, erepsin and lipase of the juice of the small intestine.

Investigations similar to those described for diastase were carried out for these enzymes. The concentrations in consecutive samples if they fell at all did not fall in comparable degree to those of diastase (*vide* Fig. 3 and Figs. 4, 5, and 6), and although the mixed juices contained usually more of each enzyme than did the pure juice, yet in 14 experiments on L.M., in the case of invertase the concentration in the mixed juice was never more than 10 times as much as in the pure juice (average 4·7 times), in that of lipase once 11 times (average 3·2 times), and in the case of erepsin only once as much as 5 times (average 2·9 times). Similarly in the same number of
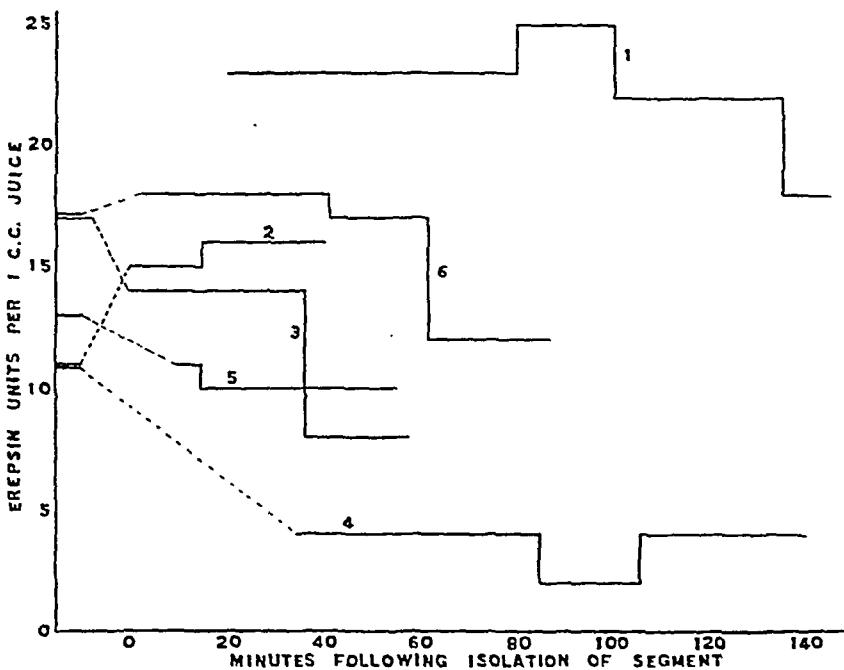
Fig. 5. The erepsin concentrations in the juice aspirated after isolating a segment of small intestine. The interrupted lines indicate periods over which no estimations of erepsin were made. Observations 1, 2, 3, and 6 upon Subject L.M.; observation 4 upon Subject I.S.E.; observation 5 upon Subject J.E.M..

experiments on I.S.E. the average values were 4, 6, 7, and 2·1 times respectively. One experiment on E.C. and one on J.E.M. gave results agreeing with these. When compared with the corresponding values for diastase, which ranged from 500 to 2000 times, these results, together with those in Table II, show incontestably that the enzymes invertase, lipase and erepsin are secreted by the mucus membrane of the small intestine in significant amounts. That the absence from the pure juice of activators present in the mixed juice did not account for the lower activity of the former was demonstrated for these enzymes in the same way as for diastase. In the case of

lipase this was surprising, as bile salts are known to augment its activity ; possibly the instability of the oil emulsion, and the lack of continuous shaking throughout the 24 hours incubation, prevented the effective action of the bile salts.

As in the case of diastase no relation was found between intestinal level, over the range studied, and enzyme concentrations in either mixed or pure juice.

It should be noted finally that the concentrations of invertase, lipase and erepsin found in the serum in 4 control experiments, namely 0, 0 to 5, and 0 to 2 units per c.c., respectively, unlike those for diastase were lower than those in the pure intestinal juice.
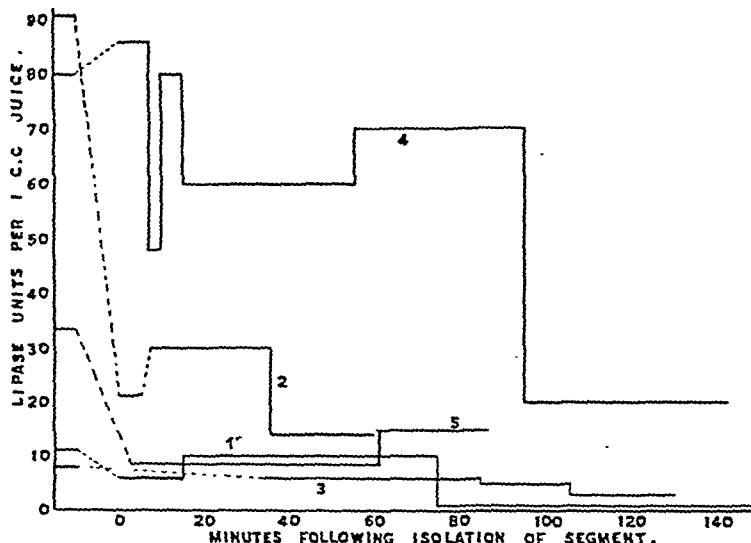

Fig. 6. The lipase concentrations in the juice aspirated after isolating a segment of small intestine. The interrupted lines indicate periods over which no estimations of lipase were made. Observations 1, 2, and 5 upon Subject L.M. ; observation 3 upon Subject I.S.E. ; observation 4 upon Subject J.E.M..

The lactase of the juice of the small intestine.

Qualitative tests for lactase were made on the pure intestinal juice of 6 subjects (14 observations). It was demonstrated in each of 9 observations on 5 of these subjects (L.M., H.E.O., J.E.M., E.C., J.W.I.). In the case of I.S.E. it was found in relatively low concentration in 3 determinations, but not in 2 others. The washing of the segment was not always equally complete, but no constant relation between this and the activity of the juice was found. Lactase was demonstrated in specimens believed to be uncontaminated, and having low diastatic activities. Therefore lactase may be accepted as a constituent of the pure juice.

SUMMARY.

1.   The diastase of the pure secretion of the small intestine is, in man, insignificant in amount, and forms at most a very small proportion of the diastase present in the mixed intestinal juices.   Thus this diastase can play only a very small rôle in the total breakdown of starch in the small intestine. Indeed it is probable that diastase is not a true secretion of the human small intestine.

2.   The invertase, erepsin and lipase of the pure juice provide a significant proportion of these enzymes, as they are found in the mixed intestinal juices, and appear to be true intestinal secretions.

3.   Lactase has been demonstrated in the intestinal juice of 6 men, and is a true secretion of the small intestine.

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INVESTIGATIONS OF THE FUNCTIONS OF THE SMALL INTESTINE IN MAN BY INTESTINAL INTUBATION.

PART 3.—FACTORS INFLUENCING THE SECRETION OF JUICE BY THE SMALL INTESTINE.

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IN dogs with Thiry fistulæ of the small intestine no secretion of intestinal juice occurs in the true resting state (Pawlow (20)), apart from a slight periodic secretion lasting a few minutes at intervals of some hours (Boldyreff (3)). Mechanical stimulation readily induces a flow of juice, the response being strictly local in character. Similarly many and various chemical substances have been found to act as efficient local stimuli, e.g., calomel, hydrochloric acid, gastric and pancreatic juices, and solutions of carbohydrates, peptone and magnesium sulphate (Röhmann (22); London (11); Babkin (1)). Section of the nerve supply to the segment leads to a continuous secretion apart from any local stimulation (Moreau (13)). In the majority of cases no effect on the secretion from the segment was found to follow oral administration of food (Pawlow (20); Babkin (1)). Some investigators found a decrease in output (Brestkin and Sawitch (4); Oppenheimer (16); Ravdin (21); Cajori (5)). Rarely, and at high intestinal levels only, an increase was found, especially after giving fats (Glinski (8)). Sawitch (23) demonstrated an increase to occur after food when the segment studied was denervated. Nasset, Pierce, and Murlin (15) found an increased secretion from jejunal transplants after food, but consistently only after denervation of the transplant. Sawitch (23) suggested that there is a normal nervous inhibition of intestinal secretion from which the gland cells are liberated by local mechanical or chemical stimulation. Babkin (1) supports this conclusion.

With continued mechanical stimulation there is a progressive fall in enzyme concentration in the juice secreted. Sawitch found a specific effect

for locally applied pancreatic juice in re-establishing the output of entero-kinase in such cases, and others have found analogous effects for pepsin and trypsin in the case of erepsin, for starch and pancreatic juice in the case of diastase, and for pancreatic juice in the case of lipase (*vide* Babkin (1)). Mechanical stimulation has been considered capable only of liberating enzymes from stores preformed in the mucosal cells, new formation depending on specific chemico-hormonal effects associated with food, and possibly with the secretion of pancreatic juice.   Apart from the exceptional case reported

TABLE I.

The effect on the volume, total enzyme output and enzyme concentration of the juice secreted by an isolated segment of small intestine of mechanical stimulation due to the pressure of the inflated balloons.

Observation.	Subject.	Segment depth in cm.	Inflation pressure of the balloons in cm. of water.	Secretion time of sample, in min. from beginning.	Rate of secretion in c.c. per 15 min..	Invertase units per 1 c.c. juice.	Invertase units per min..	Lipase units per 1 c.c. juice.	Lipase units per min..	Erepsin units per 1 c.c. juice.	Erepsin units per min..
1	L.M.	120	48	0 to 6	18·0	16	19	21	25·0	14	17·0
				6 to 8	18·0	9	11	—	—	—	—
				8 to 37	5·0	13	4	30	10·0	14	5·0
				37 to 59	4·5	7	2	14	4·0	8	2·0
2	J.E.M.	105	48	0 to 7	25·5	23	39	86	146·0	—	—
				7 to 10	24·0	23	37	48	77·0	—	—
				10 to 15	15·0	32	32	79	79·0	11	11·0
				15 to 56	2·0	68	10	61	9·0	10	1·5
				56 to 94	3·5	83	18	70	16·0	—	—
3	L.M.	120	0′ to 130′ 32·5 then 38	0 to 15	4·5	100	30	6	2·0	15	4·5
				15 to 40	5·5	215	76	10	3·5	16	5·5
				40 to 76	3·0	240	51	10	2·0	14	3·0
				76 to 150	1·0	210	17	1	trace	15	1·0
				150 to 169	6·5	140	60	4	2·0	11	5·0
				169 to 194	5·5	40	15	7	3·0	5	2·0
				194 to 210	3·0	18	3	—	—	—	—
4	I.S.E.	100	35	80 to 105	2·5	63	11	24	4·0	—	—
			45	105 to 125	6·5	47	21	23	10·0	—	—

by Delezenne and Frouin (6), who found stimulation of the secretion from an isolated segment of jejunum as a result of the insertion of N/10 hydrochloric acid into the duodenum, no such remote effect has been demonstrated.

Following the injection of histamine conflicting results have been obtained. Koskowski (10) found an increased volume and total enzyme output, Ravdin (21) an increased volume output, Cajori (5) an increased volume but unchanged total enzyme output.

In man investigations are scanty. Hamburger and Hekma (9) and others have verified the effect of mechanical stimulation, and Bickel and

Wagner (2) that of calomel. Orbéli and Sawitch (17), investigating the cæcal secretion, verified the fall of enterokinase secretion with continued mechanical stimulation and its specific restoration by locally applied pancreatic juice or oral administration of food, but did not find any similar fall in the case of erepsin. They demonstrated also local stimulation by amino-acid solutions. Démant (7) and Nagano (14) found an increased secretion of small intestinal juice in the afternoon, possibly as a result of the mid-day meal, but did not investigate this relationship further. Tubby and Manning (24) could find no relationship between variation in flow and meals. Orbéli and Sawitch (17) found no increase in volume or erepsin secretion in the large-intestinal juice with food, but found an increase in enterokinase secretion. Bickel and Wagner (2) demonstrated marked changes in small intestinal secretion (a) an immediate increase, 2 to 5 minutes after food, lasting 3 to 4 hours and considered psychic in origin, (b) a fall again to a low level for 1 to 2 hours, and (c) a second rise lasting about 2 hours, believed to be chemico-hormonal in origin.

The present investigation has attempted to obtain further data for man. The method of intestinal intubation, and the analytical methods used, have already been described (Owles (18 and 19)). The data obtained for diastase are presented in the tables throughout, but are considered separately from those on invertase, lipase and erepsin in a final section.

The effect of mechanical and chemical stimuli applied directly to the small intestine on the secretion of its juice.

The method of intestinal intubation does not permit investigation in the complete absence of any local mechanical stimulation. Following the inflation of the balloons there was a gradual fall from the initial values for both volume and total enzyme output, and often for enzyme concentration, in the juice secreted by the isolated segment, (Table I, Obs. 1 and 2). In these examples, which are selected, the possibility that this fall was due to the washing out of residual mixed juice could be excluded on account of the relative enzyme concentrations of the initial juice and that collected later, and the initial high values were probably due to stimulation resulting from the inflation of the balloons. This is supported by the fact that (Table I; Obs. 3 and 4) when the enzyme output had fallen to a low and relatively steady level a further increase in balloon pressure resulted in a secondary rise in output. If air was run in and out of the segment during the collection of the samples (Table II, Obs. 1, 2 and 4) stimulation both of volume output, and of total output of invertase, lipase and erepsin occurred. Similar slight stimulation of enzyme secretion occurred if normal saline was inserted (Table II, Obs. 3, 4 and 5), but more if it was run in and out of the segment (Table II, Obs. 3), or if air was run in and out as well (Table II, Obs. 5). The stimulating effect of air was of the same order as that of saline, and there was no evidence of any effect of the latter other than a purely mechanical one.

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TABLE II.

The effect on the volume and total enzyme output of the juice secreted by an isolated segment of small intestine of the insertion into the segment of various substances.

Observation.	Subject.	Segment depth in cm..	Nature of sample and of substance inserted into the isolated segment.	Duration of sample in min..	Rate of secretion in c.c. per 15 min..	Invertase units per min..	Lipase units per min..	Erepsin units per min..	Diastase units per min..
1	L.M.	120	Resting juice	14	5·0	10·0	1·5	2·5	
			Resting juice	72	2·5	5·0	0·5	1·5	} < 0·5
			Air run in and out of segment	15	8·0	47·0	1·0	9·0	
			Air run in and out of segment	15	6·5	25·0	2·0	5·5	
2	L.M.	160	Resting juice	32	4·0	17·0	0·3	2·5	} < 0·5
			Air run in and out of segment	10	9·0	36·0	0·5	6·0	
3	L.M.	160	Resting juice	20	7·0	20·0	2·5	3·5	} 0·5
			Resting juice	20	4·5	20·0	0·5	3·0	
			20 c.c. saline inserted	10	—	22·0	1·0	4·5	< 1·5
			20 c.c. saline run in and out	10	—	41·0	1·0	6·0	< 1·5
			10 c.c. 10% MgSO$_4$ soln. inserted	10	15·0	30·0	1·0	3·5	< 1·5
4	I.S.E.	120	Resting juice	33	5·5	10·0	2·0	0·5	
			Resting juice	13	6·5	11·0	2·0	2·0	
			Resting juice	13	8·0	14·0	2·5	2·5	
			Air run in and out of segment for last 3 min..	16	6·0	20·0	1·5	3·0	
			Air run in and out of segment	18	8·5	21·0	3·0	4·0	} < 2·0
			Air run in and out of segment	15	9·0	20·0	2·5	3·5	
			20 c.c. saline inserted	10	—	15·0	5·0	5·0	≪ 1·7
5	I.S.E.	125	Resting juice	18	5·0	9·5	6·0	3·0	
			Resting juice	27	8·0	13·0	11·0	4·5	< 1·5
			30 c.c. saline in 10 min., then air run in and out.	20	—	18·0	25·0	5·5	
			30 c.c. saline inserted	15	—	8·0	14·0	3·0	
			20 c.c. 2% glucose in 10 min., then saline wash-outs	15	—	7·0	13·0	4·0	
			20 c.c. 10% glucose in 11 min., then saline wash-outs	26	—	3·0	3·0	2·0	< 0·6
			20 c.c. 10% sucrose in 10 min., then saline wash-outs	28	—	6·5	5·0	2·0	
			20 c.c. boiled mixed juice inserted	10	—	11·0	8·5	5·5	4·0
6	I.S.E.	135	Resting juice	20	9·0	9·0	3·0	2·0	< 0·5
			20 c.c. 10% glucose solution	10	—	0·0	0·0	4·5	< 1·5

TABLE II—*continued.*

Observation.	Subject.	Segment depth in cm..	Nature of sample and of substance inserted into the isolated segment.	Duration of sample in min..	Rate of secretion in c.c. per 15 min..	Invertase units per min..	Lipase units per min..	Erepsin units per min..	Diastase units per min..
7	I.S.E.	140	Resting juice	18	7·5	11·5	3·5	6·0	
			Resting juice	16	8·5	18·5	1·5	10·0	
			20 c.c. 10% MgSO₄ solution	10	18·0	49·5	11·5	46·0	
8	L.M.	120	Resting juice	25	9·5	22·0	26·0	6·0	36·0
			Resting juice	15	6·5	28·0	16·5	—	35·0
			10 c.c. 10% MgSO₄ solution	5	30·0	50·0	26·0	26·0	1·5
9	L.M.	135	20 c.c. 2% starch solution	17	—	22·0	37·0	2·5	3·0
10	I.S.E.	130	Resting juice	27	10·5	31·0	5·5	12·5	—
			20 c.c. N/10 HCl inserted for 5 min., then consecutive samples collected	7	25·5	0·0	0·0	0·0	0·0
				11	11·0	10·5	4·5	15·5	—
				21	8·5	12·0	5·5	5·5	1·0
				37	8·5	12·0	5·5	10·0	—
11	L.M.	140	Resting juice	15	8·5	31·0	2·0	5·5	—
			25 c.c. N/10 HCl inserted for 5 min., then consecutive samples collected	6	37·5	0·0	0·0	0·0	—
				4	16·7	22·0	—	8·0	—
				27	6·8	10·0	1·5	4·5	—
				18	10·7	15·0	—	3·0	—
				10	10·0	16·0	—	3·5	—
12	L.M.	130	Resting juice	25	4·5	26·0	4·5	3·5	27·5
			30 c.c. mixed juice inserted for 5 min., then consecutive samples collected	15	—	—	—	—	—
				57	3·0	22·0	4·0	3·0	22·0
				33	2·5	23·0	4·0	3·0	21·0
				55	2·5	20·0	3·0	2·5	16·5
13	L.M.	125	Resting juice	15	4·0	—	—	—	—
			Resting juice	15	3·0	—	—	—	—
			25 c.c. mixed juice inserted for first 3 min..	18	1·5	—	—	—	—

Hypertonic magnesium sulphate solutions inserted into the segment induced a marked increase in volume output of juice, and an increased secretion of mucus, both effects being in evidence on introducing any irritant, such as distilled water, solutions of iron salts, or N/20 hydrochloric acid. In 5 minutes 10 c.c. of 10% MgSO₄ solution were diluted to approximately 20 c.c. (2 observations), 20 c.c. to 35 c.c. (5 observations), 10 c.c. of 20% solution to 40 c.c. (2 observations), and in 10 minutes, 10 c.c. of 30% solution to 70 c.c. (1 observation). Total enzyme output was also augmented (Table II, Obs. 3, 7 and 8), though usually to a less degree than was volume output. No persistence of the stimulatory effect was found in one observation 5 minutes after washing out the magnesium sulphate solution.

N/20 hydrocholoric acid introduced into the segment irreversibly inactivated all the enzymes, but was rapidly diluted and neutralized; for example, 10 c.c. N/10 hydrochloric acid were diluted in 2 experiments to 40 c.c. and 30 c.c., in 3 and 5 minutes, with final total acidities equivalent to 2 c.c. and 3 c.c. N/10 acid, respectively; 25 c.c. and 20 c.c. N/20 acid were diluted in 5 minutes to 45 c.c. and 40 c.c. respectively with a final acidity in the latter case equivalent to 3·5 c.c. acid. It was possible therefore that the stimulating effect might persist beyond the time at which the acid could inactivate any enzymes secreted, but this was not found to be so (Table II, Obs. 10 and 11), and no increase in active enzyme calculated over the whole period occurred. The local action of the acid of the gastric contents therefore cannot be a normal physiological stimulus to secretion of intestinal invertase, lipase and erepsin. Similarly no stimulating effect was found for the local action of boiled mixed intestinal juice, containing bile salts (Table II, obs. 5), or for unboiled active mixed juice (Table II, Obs. 12 and 13) of high diastatic activity, and so presumably containing pancreatic juice (19).

Such few observations as were made for carbohydrate solutions showed either no stimulation, or no greater stimulation than corresponded with the mechanical effect as seen on insertion of air or normal saline (Table II, Obs. 5, 2% glucose solution; Obs. 5 and 6, 10% glucose solution; Obs. 5, 10% sucrose solution). Following the insertion of 2% starch solution (Table II, Obs. 9) the enzyme output was within the normal range. In one observation the insertion into the segment after only slight preliminary washing with saline of two 30 c.c. volumes of 1·3% starch solution, for 15 minutes each, followed by 30 c.c. of 0·3% glucose solution for 25 minutes, failed to check the fall of diastase concentration and total output normally seen with saline alone under such circumstances (Owles (19)).

Colic developed during the course of some experiments (Table III), and there was a simultaneous increase in volume and total enzyme output of the juice of like order to that above. The colic was probably due to the pressure of the balloons, and was associated with increased tonus and motor activity of the intestine, as evidenced by movements in the air-water reservoirs used for inflation of the balloons.

TABLE III.

Variations in the volume, total enzyme output, and enzyme concentrations, of the juice secreted by an isolated segment of small intestine, occurring in association with the development of intestinal colic.

Observation.	Subject.	Segment depth in c.m.	Duration of sample in min.		Rate of flow in c.c. per 15 min.	Invertase units per 1 c.c. juice.	Invertase units per min.	Lipase units per 1 c.c. juice.	Lipase units per min.	Erepsin units per 1 c.c. juice.	Erepsin units per min.	Diastase units per 1 c.c. juice.	Diastase units per min.
1	E.C.	125	23		8.0	50.0	26.0	6.0	3.0	25.0	13.0	90	50
			10	(colic)	13.5	56.0	51.0	8.0	7.0	18.0	16.0	—	—
			45		4.0	83.0	21.0	23.0	6.0	22.0	5.5	—	—
2	L.M.	135	33		4.5	47.0	14.0	—	—	23.0	7.5	850	255
			40		4.0	48.0	12.0	—	—	25.0	6.5	150	39
			20	(colic)	7.5	45.0	23.0	—	—	22.0	11.0	60	31
			10		3.5	—	—	—	—	18.0	4.0	34	8
			6		3.5	56.0	13.0	—	—	—	—	22	5
3	L.M.	140	20	(colic)	7.5	29.0	22.0	13.0	10.0	6.5	5.0	—	—
			20	(colic)	7.5	31.0	23.5	10.0	7.5	5.0	4.0	—	—
			14		10.0	14.0	9.5	7.0	4.5	6.0	4.0	—	—
4	I.S.E.	100	25		2.5	63.0	11.0	24.0	4.0	—	—	—	—
			22	(colic)	6.5	47.0	19.5	22.0	9.5	28.0	11.5	10	5
5	I.S.E.	200	26		7.0	15.0	7.0	6.0	2.5	4.0	2.0	10	5
			6	(colic)	11.5	14.0	11.0	6.0	4.5	3.0	2.5	—	—
			12		8.0	12.0	6.0	6.0	3.0	4.0	2.0	—	—
6	I.S.E.	210	37		5.0	34.0	11.0	23.0	7.5	10.5	3.5	—	—
			24	(colic)	8.0	30.0	17.0	20.0	11.0	10.0	5.5	—	—
			24		8.5	27.0	16.0	30.0	17.0	11.0	6.5	—	—
			34		6.0	28.0	11.0	23.0	9.0	12.0	4.5	1	0.5

In every case the individual enzymes, invertase, lipase and erepsin were similarly affected. Diastase will be considered separately later. Enzyme output kept parallel to volume changes rather than to enzyme concentrations of the juice.

If air (Table IV, Obs. 1, 5 and 6), or saline (Table IV, Obs. 2, 4 and 6) was run in and out of the intestine above the segment a similar stimulation of the juice secreted by the isolated segment was seen, of like order in the two cases. 5% glucose solutions (Table IV, Obs. 2 and 3), and 15% MgSO₄ solution (Table IV, Obs. 3) similarly introduced produced a corresponding degree of stimulation. N/20 hydrochloric acid (Table IV, Obs. 2) produced no significant stimulation. In this series of observations the solutions were run directly into the 2nd part of the duodenum, since it seemed probable that in this way any specific effects would be most readily demonstrated, but no such effect in excess of that with air or saline was found.

TABLE IV.

The effect on the volume, total enzyme output and enzyme concentrations of the juice secreted by an isolated segment of small intestine of the introduction of various substances into the intestine above the segment.

Observation.	Subject.	Segment depth in cm..	Substances inserted into the intestines above the isolated segment.	Secretion time of sample in min..	Rate of secretion in c.c. per 15 min..	Invertase units per 1c.c. juice.	Invertase units per min..	Lipase units per 1c.c. juice.	Lipase units per min..	Erepsin units per 1c.c. juice.	Erepsin units per min..	Diastase units per 1c.c. juice.
1	L.M.	105		0 to 9	7·5	32	16·0	8·0	4·0	9·0	4·5	—
				9 to 20	6·5	35	15·0	8·0	3·5	9·0	4·0	—
				20 to 66	1·5	71	8·0	4·0	0·5	15·0	1·5	—
			71' to 81': 60 c.c. air run in and out.	66 to 80	5·5	104	37·0	8·0	3·0	23·0	8·0	—
				80 to 100	5·0	68	23·0	21·0	7·0	14·0	5·0	—
				100 to 120	4·5	83	24·0	1·0	0·5	14·0	4·0	—
2	L.M.	105	? diluted with saline from initial washing. 40' to 50': 50c.c. saline in duodenum	0 to 44	5·0	25	8·5	6·0	2·0	8·0	3·0	—
				44 to 54	10·0	32	32·0	7·0	7·0	11·0	11·0	—
				54 to 63	8·5	45	26·0	8·0	4·5	—	—	—
			62' to 72': 60 c.c. N/20 HCl in duodenum. 97' to 102': 30 c.c. 5% glucose in duodenum.	63 to 100	4·5	43	13·5	6·0	2·0	11·0	3·5	—
				100 to 116	10·0	42	28·0	4·0	2·5	11·0	7·5	—
				116 to 132	4·5	74	23·0	7·0	2·0	13·0	4·0	—
3	L.M.	135		0 to 34	8·5	14	8·0	5·0	3·0	4·0	2·5	—
				34 to 47	7·5	20	10·0	4·0	2·0	4·5	2·0	—
				47 to 66	6·0	18	7·0	5·5	2·0	5·5	2·0	—
			65' to 75'; 15 c.c. 5% glucose in duodenum	66 to 85	9·0	18	10·5	4·0	2·5	4·5	2·5	1·5
				85 to 97	9·5	18	11·5	3·0	2·0	4·5	3·0	—
			105' to 115': 15 c.c. 15% MgSO₄ in duodenum.	97 to 110	10·0	18	12·0	3·0	2·0	4·5	3·0	—
4	L.M.	160	80 c.c. saline inserted at 27': Reaspirated from 30' to 55'	0 to 28	4·0	57	15·0	2·5	0·5	10·0	2·5	
				28 to 43	5·5	56	21·0	1·0	0·5	8·0	3·0	} 1·5
				43 to 75	4·0	66	16·5	1·0	0·5	10·0	2·5	
5	I.S.E.	200		0 to 71	5·5	21	8·0	7·0	2·5	5·0	2·0	—
				71 to 88	6·5	16	7·0	6·0	2·5	5·0	2·0	<2·0
			100' to 120': 60 to 100 c.c. air run in and out	88 to 105	7·0	15	7·0	6·0	3·0	4·0	2·0	<2·0
				105 to 122	9·5	15	10·0	6·0	4·0	3·0	2·0	—
				122 to 148	7·0	15	7·0	6·0	2·5	4·0	2·0	10·0
6	I.S.E.	215		0 to 72	6·5	20	9·0	6·0	2·5	3·0	1·5	9·0
				72 to 96	6·0	17	7·0	3·0	1·0	4·0	1·5	1·0
			96' to 111': 40 c.c. air in and out 111' to 136': 50 c.c. saline in and out	96 to 112	7·0	12	6·0	5·0	2·5	5·0	2·5	1·0
				112 to 132	9·5	14	9·0	5·0	3·0	3·0	2·0	<1·0
				132 to 146	6·5	14	6·0	0·0	0·0	4·0	2·0	<1·0
				146 to 169	5·0	21	7·0	4·0	1·5	4·0	1·5	<1·0
				169 to 194	9·5	16	3·0	4·0	0·5	5·0	1·0	<1·0

The tendency for a fall in enzyme output to occur with continuance of mechanical stimulation (Table I, Obs. 1, 2 and 3 ; Table II, Obs. 1 and 4) shows that a continued stimulus soon becomes less effectual. This tendency was seen up to the longest periods followed, 5 hours from initial inflation of the balloons, and 3 hours from the termination of any washing of the segment. In dogs this continued fall has been attributed to the gradual exhaustion of an enzyme store from the mucosal cells, but there would seem no good reason for separating it from the initial more rapid fall, which is undoubtedly due to adaptation to a continued stimulus, especially as food administered orally, and entering the intestine above the segment during the observation, did not check the fall (Table V, Obs. 7), nor did the insertion into the segment of mixed juice, presumably containing pancreatic juice (Table II, Obs. 12).

The results obtained therefore agree with those on animals in establishing mechanical stimuli as effective local augmentors of intestinal secretion, but differ in that they demonstrate a spread of the response to neighbouring parts of the intestine. A possible explanation of this difference is that the operative procedure involved in the establishment of a Thiry fistula necessitates the transection of any local nerve plexuses. Such nerve connections are maintained intact in the present method of investigation, and are the probable means of transmitting stimuli from neighbouring parts of the intestine. No specific action was demonstrated, either as regards their local or remote effects, for the various solutions studied.

The effect of orally administering various substances on the secretion of juice by an isolated segment of small intestine.

Table V gives the results of experiments in which this effect was investigated, the substances administered being reaspirated throughout from a point immediately above the isolated segment. Stimulation of volume output of juice, and of total output of invertase, lipase and erepsin was demonstrated following the drinking of water (Obs. 3, 4 and 6), the administration of eggs and milk (Obs. 1, 2 and 8), of glucose solution (Obs. 5), of ferrous sulphate solution (Obs. 11), and of ferric ammonium citrate solution (Obs. 12). In one instance on I.S.E. (Obs. 9), the drinking of glucose solution resulted in little, and in another (Obs. 10) in no stimulation. No consistent differences were noted for relative changes in output of the individual enzymes.

Table VI contains such observations as are available, in which it is possible to compare the responses to similar stimuli at different intestinal levels in the same individual. It is clear that there is a tendency to a greater response at higher intestinal levels, this agreeing with the scanty observations of former workers. Taking this fact into account there is no evidence forthcoming from the above results for any specific effect for food substances in augmenting the secretion from the segment, comparable degrees of

TABLE V.

The effect on the volume, total enzyme output and enzyme concentrations of the juice secreted by an isolated segment of small intestine of oral administration of various substances. The asterisk indicates the first sample of material aspirated just proximal to the isolated segment which contained the substance administered.

Observation.	Subject.	Segment depth in cm.	Substance administered orally.	Secretion time in min. of sample, relative to administration of food, etc.	Juice from segment in c.c. per 15 min..	Juice above segment in c.c. per 15 min..	Diastase units per 1 c.c. juice	Invertase units per 1 c.c. juice.	Invertase units per min.	Lipase units per 1 c.c. juice.	Lipase units per min.	Erepsin units per 1 c.c. juice.	Erepsin units per min..
1	E.C.	125	2 eggs at 0'	— 95 to — 66	8·5	1	—	43	24·0	6·0	3·5	11·0	6·0
				— 66 to — 43	8·0	0	—	50	26·0	6·0	3·0	25·0	13·0
			Colic from —50' to —30'	— 43 to — 32	13·5	14	—	56	51·0	8·0	7·0	18·0	16·0
				— 32 to + 12	4·0	1	—	83	21·0	23·0	6·0	22·0	5·5
				+ 12 to + 26	9·0	3*	—	73	45·0	37·0	23·0	14·0	8·5
				+ 26 to + 42	6·5	13	—	81	35·0	23·0	10·0	22·0	9·5
				+ 42 to + 55	9·0	26	—	}58	36·0	}17·0	10·5	}24·0	12·5
				+ 55 to + 65	7·5	9	—		29·0		8·5		10·0
2	L.M.	130	1 egg and 200 c.c. milk at 0'	—150 to —143	3·0	0	800	110	22·0	19·0	4·0	14·0	3·0
				—143 to — 60	2·5	7	500	127	23·0	21·0	4·0	16·0	3·0
				— 60 to — 5	2·5	0	250	110	20·0	17·0	3·0	13·0	2·5
				— 5 to + 14	6·5	75*	90	126	54·0	20·0	8·5	14·0	6·0
				+ 14 to + 27	11·0	100	20	62	45·0	16·0	12·0	8·0	6·0
				+ 27 to + 42	6·0	—	15	192	74·0	—	—	14·0	5·5
3	L.M.	135	300 c.c. water at 0'	— 81 to — 57	5·5	2	—	103	39·0	1·5	0·5	9·0	3·5
				— 57 to — 33	8·0	2	—	90	49·0	1·5	1·0	10·5	5·5
				— 33 to — 22	14·5	44	—	85	83·0	3·0	3·0	10·5	10·5
				— 22 to + 0	6·0	8	—	63	26·0	0·5	0·5	7·5	3·0
				0 to + 9	22·0	19*	—	64	94·0	1·5	2·0	7·5	11·0
				+ 9 to + 33	19·0	40	<0·7	44	56·0	1·5	2·0	6·5	8·5
4	L.M.	150	350 c.c. water at 0'	— 91 to — 70	4·0	2	—	68	19·0	15·0	4·0	8·5	2·5
				— 70 to — 26	4·5	16	<1·0	51	15·0	5·0	1·5	9·5	3·0
				— 26 to + 11	5·5	21*	<1·0	54	19·0	3·5	1·5	8·5	3·0
			2 eggs and 250 c.c. milk at 80'	+ 11 to + 43	5·5	50	—	47	18·0	6·0	2·5	8·5	3·5
				+ 43 to + 86	3·5	5	—	56	12·5	4·5	1·0	8·0	2·0
				+ 86 to +109	6·5	12*	—	—	—	—	—	—	—
5	L.M.	155	100 c.c. 10% glucose solution at 0'	— 39 to + 10	4·5	16	—	57	16·5	39·0	11·0	14·0	4·0
				+ 10 to + 17	17·5	38*	—	35	41·0	23·0	27·0	14·0	16·5
				+ 17 to + 30	13·0	50	—	51	45·0	31·0	27·0	27·0	24·0
6	L.M.	160	500 c.c. water at 0'	— 48 to — 27	7·0	10	}1·5	44	20·0	5·0	2·5	8·0	3·5
				— 27 to — 4	4·5	7		65	20·0	2·0	0·5	9·0	3·0
				— 4 to + 24	5·5	3*		72	27·0	2·0	1·0	9·0	3·5
				+ 24 to + 34	6·0	20		—	—	—	—	—	—

TABLE V—*continued.*

Observation.	Subject.	Segment depth in cm..	Substance administered orally.	Secretion time in min., of sample, relative to administration of food, etc.	Juice from segment in c.c. per 15 min..	Juice above segment in c.c. per 15 min..	Diastase units per 1 c.c. juice	Invertase units per 1 c.c. juice.	Invertase units per min..	Lipase units per 1 c.c. juice.	Lipase units per min..	Eropsin units per 1 c.c. juice.	Eropsin units per min..
7	L.M.	160	2 eggs and 300 c.c. Milk at 0'	− 42 to − 10	4·0	26		66	16·5	1·0	0·5	10·0	2·5
				− 10 to 0	9·0	6		60	36·0	1·0	0·5	10·0	6·0
				− 0 to + 15	8·5	5	} 1·5	58	33·5	3·0	1·5	10·0	6·0
				+ 15 to + 35	8·5	41*		51	29·5	1·0	0·5	8·0	4·5
			Air run in and out from —10 on	+ 35 to + 50	7·0	28		47	22·5	1·0	0·5	7·0	3·5
8	I.S.E.	105	2 eggs, cereal and cream at 0'	− 97 to − 52	2·5	8	4·5	153	24·0	39·0	6·5	14·0	2·0
				− 52 to + 6	2·0	5	4·0	212	26·5	45·0	5·5	18·0	2·0
				+ 6 to + 34	4·5	47*	2·0	196	58·5	45·0	12·0	18·0	5·5
				+ 34 to + 47	7·0	55	2·0	170	79·5	31·0	14·5	17·0	8·0
				+ 47 to + 52	7·5	16	1·5	} 165	83·0	} 25·0	12·5	} 17·0	8·5
				+ 52 to + 88	2·0	6	3·0		23·5		3·5		2·5
9	I.S.E.	135	100 c.c. 10% glucose in orange juice, at 0'	− 50 to − 23	6·5	13	<1·5	23	10·0	21·0	9·5	8·0	3·5
				− 23 to − 0	7·5	17	<1·5	21	11·0	18·0	9·0	6·0	3·0
				− 0 to + 18	9·5	33*	<1·0	17	10·5	16·0	10·0	4·0	2·5
				+ 18 to + 40	7·5	24	1·0	17	8·5	13·0	6·0	4·0	2·0
				+ 40 to + 62	9·5	15	1·0	17	10·5	8·0	5·0	3·5	2·0
				+ 62 to + 77	10·5	19	<1·0	15	10·5	3·0	2·0	3·0	2·0
				+ 77 to + 96	9·0	30	<1·0	15	9·0	4·0	2·5	3·0	2·0
10	I.S.E.	150	200 c.c. 25% glucose in orange juice	− 27 to + 0	8·0	40	2·0	14	7·5	11·0	6·0	6·0	3·5
				0 to + 28	7·5	102*	—	12	6·0	11·0	5·5	6·0	3·0
				+ 28 to + 40	7·5	74	—	12	6·0	10·0	5·0	9·0	5·0
				+ 40 to + 60	6·0	49	<1·5	17	7·0	13·0	5·0	8·0	3·0
11	I.S.E.	195	80 c.c. FeSO₄ soln. (= ½g. iron) at 0'	− 37 to 0	3·0	—	—	32	7·0	22·0	4·5	14·0	3·0
				0 to + 22	5·5	—*	—	30	11·0	44·0	16·0	27·0	10·0
12	I.S.E.	210	80 c.c. ferric ammonium citrate soln. (= ½g. iron) at 0'	− 60 to − 27	3·5	16	—	37	9·0	23·0	8·5	16·0	4·0
				− 27 to + 12	6·0	19*	—	29	11·0	31·0	12·0	8·0	3·5
				+ 12 to + 40	5·5	68	—	40	15·0	25·0	10·0	17·0	6·5
				+ 40 to + 77	5·0	15	—	34	11·0	23·0	7·5	11·0	3·5
				+ 77 to +101	8·0	—	—	30	17·0	20·0	11·0	10·0	5·5
				+101 to +125	8·5	10	—	27	15·5	30·0	17·0	11·0	6·5
			Colic + 75' to + 95'	+125 to +160	6·0		1·0	28	11·0	23·0	8·5	12·0	4·5

TABLE VI.

The total enzyme output in the juice secreted by isolated 10 cm. segments at different levels of small intestine in the same individual, in response to similar stimuli.

Subject.	Segment depth in cm..	Nature of stimulus to secretion.	Enzyme units per minute.		
			Invertase	Lipase	Erepsin
L.M.	120	Air run in and out of isolated segment.	47·0	1·0	9·0
	160	Air run in and out of isolated segment.	36·0	0·5	6·0
L.M.	105	50 c.c. saline inserted above isolated segment.	32·0	7·0	11·0
	160	80 c.c. saline inserted above isolated segment.	21·0	0·5	3·0
L.M.	135	300 c.c. water by mouth.	94·0	2·0	11·0
	150	350 c.c. water by mouth.	19·0	2·5	3·5
	160	500 c.c. water by mouth.	27·0	1·0	3·5
L.M.	130	Egg in milk by mouth.	54·0	12·0	6·0
	160	The same, with local stimulation by air.	36·0	1·5	6·0
L.M.	105	30 c.c. 5% glucose solution inserted above segment.	28·0	2·5	7·5
	135	15 c.c. 5% glucose solution inserted above segment.	11·5	2·5	3·0
	135	15 c.c. 10% MgSO₄ solution inserted above segment.	12·0	2·0	3·0
I.S.E.	100	Colic.	19·5	9·5	11·5
	200	Colic.	11·0	4·5	2·5
	210	Colic.	17·0	17·0	6·5
I.S.E.	135	100 c.c. 10% glucose by mouth.	17·0	10·0	2·5
	150	200 c.c. 25% glucose by mouth.	7·0	5·5	5·0

stimulation occurring with all the substances investigated. In Observation 7 administration of a meal of eggs and milk to L.M. neither increased further the secretion induced by local mechanical stimulation with air, nor prevented the fall in enzyme output with continued stimulation. In Observation 1, colic was associated with a similar degree of stimulation to that induced by food, in each case marked. In Observation 3, a spontaneous flow of juice down the intestine above the segment (-33 to -22 min.) was associated with an increase in output from the segment comparable to that occurring later in the same experiment on administration of water by mouth, and in Observation 2 of egg and milk. Under the conditions of the experiments food rapidly entered the intestine from the stomach, and some usually reached a level just above the segment within 10 minutes. It was not possible therefore to study any effect before this occurred. An increase in flow from the segment might precede by 1 to 10 minutes the arrival of food substances above the segment, but could always be taken as a definite indication of the occurrence of the latter within that period, and probably

TABLE VII.

The effect of the injection of 0·5 mg. histamine phosphate on the volume, total enzyme output, and enzyme concentrations of the juice secreted by an isolated segment of intestine, and on the volume of juice coming down the intestine above the segment.

Observation.	Subject.	Segment depth in cm..	Secretion time of sample in min., relative to injection of histamine.	Flow from segment in c.c. per 15 min..	Flow from above segment in c.c. per 15 min.	Invertase units per 1 c.c. juice.	Invertase units per min..	Lipase units per 1 c.c. juice.	Lipase units per min..	Eropsin units per 1 c.c. juice.	Eropsin units per min..	Diastase units per 1 c.c. juice.	Diastase units per min..
1	E.C.	105	— 55 to — 5	1·5	2	238·0	26·0	17	2·0	46	5·0	9·0	1·0
			— 5 to + 41	2·0	0	172·0	22·5	17	2·5	34	4·5	4·5	0·5
			+ 41 to + 70	2·5	2	142·0	24·5	8	1·5	35	6·0	6·0	0·5
			+ 70 to + 85	5·0	14	—	—	—	—	—	—	—	—
2	J.W.I.	115	— 22 to — 10	11·5	22	19·0	17·0	7	6·0	8	7·0	7·0	6·0
			— 10 to + 3	11·0	0	23·0	16·5	6	4·5	8	6·0	5·0	3·5
			+ 3 to + 31	8·0	0	26·0	13·5	6	3·0	7	6·0	5·0	2·5
			+ 31 to + 55	10·5	6	—	—	—	—	—	—	—	—
3	I.S.E.	135	— 25 to — 7	8·5	2	30·0	17·0	25	14·0	7	4·0	0·7	0·5
			— 7 to + 17	9·0	15	33·0	20·0	27	16·5	6	3·5	<0·7	<0·5
			+ 17 to + 38	9·5	26	20·0	13·0	32	21·0	8	5·0	<0·7	<0·5
			+ 38 to + 54	10·0	5	30·0	20·0	20	13·0	11	7·0	<0·7	<0·5
			+ 54 to + 69	12·0	33	32·0	26·0	21	17·0	7	5·5	<0·7	<0·5
			+ 69 to + 93	9·0	26	20·0	12·0	17	10·0	7	4·0	<0·7	<0·5
			+ 93 to +107	13·0	22	33·0	29·0	17	15·0	8	7·0	<0·7	<0·5
4	I.S.E.	140	— 31 to — 11	8·5	9	26·5	15·0	14	8·0	6	3·5	—	—
			— 11 to + 8	7·5	10	23·0	11·5	7	3·5	13	6·0	—	—
			+ 8 to + 54	7·5	18	25·0	12·5	11	5·5	6	3·0	—	—
			+ 54 to + 85	9·5	11	27·0	17·5	10	6·5	12	7·5	—	—
			+ 85 to +109	8·5	31	29·0	16·5	10	5·5	6	3·5	0·3	0·1
			+109 to +125	8·5	57	32·5	18·5	3	1·5	18	10·0	—	—
5	I.S.E.	150	— 40 to — 15	6·0	6	37·5	15·5	10	4·0	8	3·0	—	—
			— 15 to 0	6·0	2	32·5	13·0	10	4·0	9	3·5		—
			0 to + 36	5·5	35	35·0	13·0	10	3·5	11	4·0	8·0	3·0
			+ 36 to + 51	9·5	11	19·0	12·0	11	7·0	9	5·5	—	—
			+ 51 to + 78	8·5	44	17·5	10·5	10	5·5	8	5·0	2·0	1·0
			+ 78 to +107	8·0	40	14·0	7·5	11	6·0	6	3·5	—	—
6	L.M.	125	— 15 to — 7	6·5	3	43·0	19·0	8	3·5	—	—	—	—
			— 7 to + 23	5·5	6	50·0	19·0	3	1·0	—	—	—	—
			+ 23 to + 30	3·0	25	—	—	6	1·5	—	—	—	—
			+ 30 to + 60	3·5	10	—	—	—	—	—	—	—	—
7	L.M.	195	— 30 to — 15	5·0	1	—	—	—	—	—	—	—	—
			— 15 to 0	2·5	3	—	—	—	—	—	—	—	—
			0 to + 15	2·0	0	—	—	—	—	—	—	—	—
			+ 15 to + 30	1·5	0·5	—	—	—	—	—	—	—	—
			+ 30 to + 45	0·5	0·5	—	—	—	—	—	—	—	—

only occurred when food was already entering the intestine. It was associated with increased motor activity of the intestine, and colic sometimes developed at that time (observations in which the latter occurred have been excluded from the results quoted). The possibility that the various substances administered induce their effects by producing some common stimulant cannot be excluded, but it would seem likely that were this the case greater quantitative differences in response would have been encountered. In view of the demonstration above of the similar degree of the effect of remote mechanical stimuli, therefore, it is considered that the results obtained may be explained as being associated with the mechanical and motor stimuli involved in the actual passage down the intestine of the solutions and food substances, though a chemico-hormonal mechanism in addition cannot be excluded definitely.

The effect of injecting histamine on the secretion of juice by the small intestine.

Changes in secretion from the isolated segment following the injection of 0·5 mg. histamine phosphate were never large (Table VII). There was a definite tendency for some increase of both volume and total enzyme output from the segment to occur ; this might be preceded during the first 30 minutes or so by a fall. The same in general applied to the volume of fluid coming down the intestine above the segment. Following the injection the pylorus seemed often to shut down temporarily, opening again later to admit gushes of gastric contents into the intestine, this being in agreement with the findings of Miller and Abbott (12). It is felt that the small changes in secretion from the segment which occurred were related not to any specific action of the histamine, as is the case with the gastric secretion, but to mechanical stimuli involved in the passage of the gastric contents down the intestine.

The secretion of diastase in the juice of the small intestine in response to various stimuli.

It was shown in a previous publication (19) that the pure juice of the small intestine, obtained by the method of intestinal intubation, *i.e.*, under the influence of mechanical stimulation, contains only an insignificant amount of diastase, usually 1/500 to 1/2000 that contained in the same juice mixed with pancreatic and other secretions in the proportions which normally exist in the small intestine. It was possible however that under other conditions this amount might attain significant levels. During the present investigations such an increase has been encountered in no instance, whether the diastase was studied during local or remote mechanical or chemical stimulation of the intestine, after food, or after the injection of histamine. In the case of the other enzymes studied variations in total output have tended to be associated with changes in volume flow rather than in enzyme concentration, and variations in diastase secretion of similar magnitude are

not excluded by the above results. A far greater increase than this would be necessary, however, were the diastase output to reach a significant level, and such did not occur. In view of the relatively great effect of the smallest leakage into the segment on the diastase values it was not considered profitable to follow quantitatively such small changes as possibly occurred. No evidence has been obtained therefore for any condition in which the diastase of the juice of the small intestine can be considered to play any significant part in the normal digestive processes.

SUMMARY.

1. Local mechanical stimuli are effective augmentors of the volume and total enzyme output (invertase, lipase and erepsin) of the juice of the small intestine. The response spreads beyond the actual location of the stimulus.

2. There is an increase in the volume and total enzyme output in association with colic.

3. No specific effect upon the secretion of juice by the small intestine has been found for physiological saline, carbohydrate solutions, N/10 or N/20 hydrochloric acid, bile salts, or mixed intestinal juice, whether inserted locally into the segment of intestine studied, or into remote parts of the small intestine.

4. Local irritants (*e.g.*, acid and iron salt solutions) and hypertonic solutions induce an increased output of fluid and of mucus. Hypertonic magnesium sulphate solution leads to an increase in total enzyme output. N/10 and N/20 hydrochloric acid are rapidly neutralized in the intestine; they irreversibly inactivate all the enzymes studied, and do not increase the total output of active enzyme.

5. Oral administration of water, glucose solutions, eggs and milk, and of solutions of iron salts induces an increased volume, and total enzyme, output from the segment.

6. Injection of histamine causes a temporary fall, followed in up to 30 minutes by a slight rise in volume, and in total enzyme, secretion from the segment. This is not considered specific in nature.

7. The secretion of juice in response to a given stimulus is greater at higher than at lower levels of the small intestine.

8. No evidence has been obtained for a specific chemico-hormonal mechanism, by means of which the secretion of small-intestinal juice is regulated in relation to the digestive processes.

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THE EFFECT OF LARGE DOSES OF IRON ON THE ABSORPTION

OF PHOSPHORUS.*

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IT has been shown (Brock and Diamond, 1934) that a diet which allows of normal development in young rats can be rendered rachitogenic by the addition to it of large quantities of iron. This effect was observed with ferric, ferrous and scale preparations. The amount of iron actually ingested by the rats is unknown, but the ratio of iron to phosphorus was 1·5. When, by the addition of extra dibasic sodium phosphate to the diet, this ratio was reduced to 0·8, rickets did not develop. It was concluded that the rickets was consequent on a state of phosphorus deficiency resulting from the precipitation by iron of insoluble phosphates in the intestine.

It seemed important to discover whether the large doses of iron used in clinical medicine could interfere with the assimilation of phosphorus in the human body. Mineral balance studies were, therefore, undertaken on patients with this object. Several difficulties were encountered which have an important bearing on the whole principle of mineral balance experiments, and which if ignored may lead to fallacious results. These difficulties are discussed later.

The organisation of a metabolism ward and the importance of rigid control of patients and collection of the samples have been fully dealt with by Bassett, Eldon and McCann (1931) and need not be dealt with here.

Plan of experiments.

Three-day metabolism periods were used. The patient was put on a constant weighed diet which was cooked with distilled water in aluminium vessels, and was given a few days on the diet before the collection of samples began. Millet seed was used for marking off the stool periods since carmine is obscured when the stools become black with iron. The patient was

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allowed as much distilled water as he desired to drink. In the case of an experiment starting on a Monday the following method of dividing the metabolism periods was used. After a few preliminary days on the diet, two teaspoonsful of millet seed were administered at 8 p.m. on Sunday evening. The urine for the first period was collected from 6 a.m. on Monday to 6 a.m. on Thursday. The first stool containing millet seed was discarded and subsequent stools put into the collection jar for the first period. Millet seed was administered again on Wednesday evening and the first stool containing millet seed was put into the jar for period 1 and subsequent stools into the jar for period 2. The stools in the first jar were regarded as representing the residue of the food and any drugs administered during the three days, Monday to Wednesday.

Under these conditions, even in the absence of drugs, considerable variations in the weight of mineral ash in the stools was found from period to period. (*See* Stage 1 in Table I and Fig. 1). Aperients were avoided
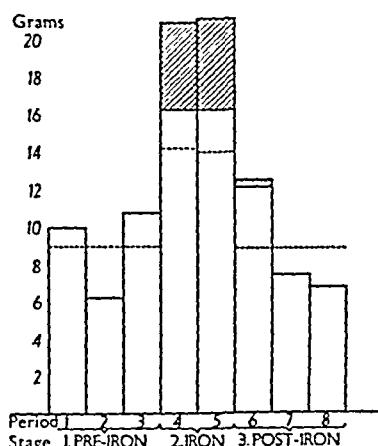
Fig. 1. Weight of mineral ash in stools as influenced by large doses of iron given in stage 2. Each column represents a three-day period, and in stages 1 and 3 the average per period is represented by a dotted line. Diagonal-hatching represents unabsorbed iron expressed as Fe. The amount of unabsorbed iron in stage 2 if expressed as Fe_2O_3 would be extended downwards to the dotted lines in that stage.

whenever possible, and when these were necessary cascara made up in distilled water was used. It was found impossible under even the most rigid supervision to obtain a constant weight of mineral ash in the stools over a consecutive series of periods. As soon as minerals, such as iron, were administered therapeutically even wider fluctuations occurred. The first source of fallacy is in this difficulty. Comparisons of the excretion of any element in the stools as between individual three- or even five-day periods mean nothing at all. Three three-day periods were found to be the minimum basis for comparison, and the weight of mineral ash should always be recorded.

TABLE I.

Case 16 B. Calcium phosphorus and iron balance for eight three-day periods. Iron and ammonium citrate administered during periods 13 and 14.

Stage	No.	No. of days	Therapy	Intake Ca	Intake P	Intake Fo Food	Intake Fo Therapy	Intake Fo Total	Stools Dry wt.	Stools Mineral ash	Ca U	Ca F	Ca T	P U	P F	P T	Fo F
1	10	3	Nil	2·13	1·32	18·0	—	18·0	55	0·05	0·35	1·89	2·24	0·93	1·01	1·93	18
	11	"	Nil	"	"	"	—	18·0	20	0·13	0·35	1·30	1·65	1·01	0·75	1·70	9·6
	12	"	Nil	"	"	"	—	18·0	53	10·75	0·31	2·30	2·61	0·84	1·28	2·12	15
2	13	"	Iron and ammon. citrate 40 grains t.i.d.	"	"	"	5375	5394	84	20·8	0·30	2·10	2·55	0·79	1·73	2·52	4040
	14	"		"	"	"	5375	5394	05	21·0	0·25	2·07	2·32	1·21	0·86	2·07	4030
3	15	"	Nil	"	"	"	—	18·0	63	12·4	0·25	2·16	2·41	0·02	1·29	2·21	203
	16	"	Nil	"	"	"	—	18·0	30	7·43	0·27	1·06	1·33	1·05	0·89	1·04	12
	17	"	Nil	"	"	"	—	18·0	00	6·70	0·31	2·04	2·35	0·95	1·05	2·00	12

Iron excretion figures in milligrams. All other excretion figures in grams. U = urine; F = faeces.
T = total (i.e., urine + faeces);

Analytical methods.

The method of preparing the stools for mineral analysis has been described elsewhere (3). By this method the mineral salts left after dry-ashing are in solution in approximately 20% hydrochloric acid. Phosphorus was determined in these solutions by the gravimetric magnesium pyrophosphate method, and calcium by the method of McCrudden (1909).

Phosphorus was determined in the urine, which was preserved by the addition of hydrochloric acid and chloroform, by the uranium acetate method (Peters and van Slyke, page 861). Calcium was determined in the urine by the method of McCrudden after the urine had been treated with ammonium persulphate (Shohl and Pedley, 1922).

Results.

Brief clinical details of the five cases studied are given in the appendix. Two of them (Cases 20 and 21) were typical cases of iron deficiency anæmia. In the other three there was no reason to suspect a state of iron deficiency.

In each case the metabolism test was divided into three stages. A fixed diet was allowed throughout the whole test. In the first stage, consisting of three or four metabolism periods, no iron was administered. During the next two or three periods large doses of iron were administered, and in the third stage, consisting of two or three periods, the iron therapy was discontinued. These three stages will be referred to as the " pre-iron," " iron," and " post-iron " stages.

In Table I the results of one case are given in full, and in Fig. 1 the weights of mineral ash for each period in the same case are shown in the form of blocks. In Table II the results of the five cases are summarised. Fig. 1 shows that the mineral ash weight is markedly increased in the stage when iron is being administered, and that the weight of unabsorbed iron, whether expressed as iron or as ferric oxide, does not account for the whole of the increase. It might be argued that the increase is merely due to intestinal hurry consequent on the commencement or iron therapy. If this were so, however, the increase should be compensated for by a reduction of the amount of mineral ash in the post-iron stage as compared with the pre-iron stage. Actually the average weight for the first and third stages, expressed by the dotted line, is equal. In Fig. 2 the average weight of mineral ash per period of each stage is expressed as a block. The five cases are illustrated separately and finally averaged. In only one case (No. 21) is there evidence of intestinal hurry in the form of a low average weight in stage 3 as compared with stage 1. In this case the iron had a laxative effect. Fig. 2 shows that in each case the administration of iron in the second stage led to an increase in the weight of mineral ash, and that this increase was in no case accounted for more than partially by the weight of unabsorbed iron.

In view of the findings previously reported on rats, the question arises whether part of this rise in mineral ash weight as a result of iron therapy can consist of phosphorus which has been precipitated as ferric phosphate and so not absorbed. In Fig. 2 the fæcal phosphorus in stages 1 and 2 is depicted for each case by transverse hatching. It is expressed as P_2O_5 because $Fe_2O_3 + P_2O_5 = 2Fe\,PO_4$. It is evident that in each case there is an increase in fæcal phosphorus in the iron stage. In Case 21 the unabsorbed iron expressed as Fe_2O_3 and the increase in fæcal phosphorus expressed as P_2O_5 account for almost the whole of the rise in mineral ash weight in the iron stage. In the other four cases, however, Fe_2O_3 and P_2O_5 together account for only a part of the increase. When the five cases are averaged it is found that the increase in mineral ash as a result of iron therapy *apart*
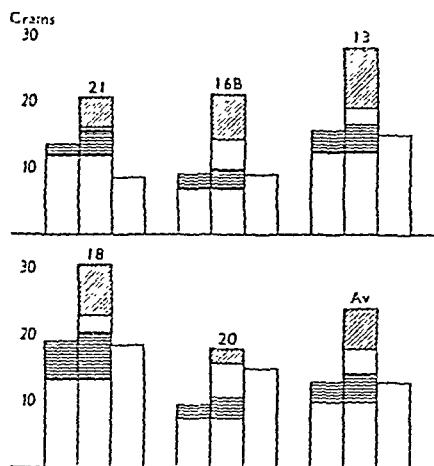
Fig. 2. The effect of large doses of iron on the weight of mineral ash in the stools per stage. Five cases individually and averaged (Av.) Diagonal hatching represents unabsorbed iron expressed as Fe_2O_3.

Transverse hatching represents phosphorus in the stools expressed as P_2O_5. The three columns in each case represent the average weight per period in the three stages.

from the weight of unabsorbed iron is 29%, (or 3·94 grams per period). This includes an average increase of 39% in the fæcal phosphorus. If the unabsorbed iron is expressed as Fe_2O_3 and the fæcal phosphorus is reckoned as P_2O_5 the unexplained increase in fæcal mineral ash amounts to an average of 2·77 grams per period. The increase is not due to calcium for the increase in fæcal calcium in the five cases averaged only 0·4% or 0·18 grams. It is usually taught that the black colour of the stool of a patient on iron therapy is due to iron sulphide, and it would be interesting to know if any part of the unexplained increase in mineral ash is due to extra sulphur which has been transformed into sulphate. Unfortunately the method of treating the stools in preparation for analysis allows of loss of sulphur, so

TABLE II.

Mineral balance figures for 5 cases.   The three stages are before, during and after administration of iron.   Figures represent averages per period of each stage.

Case No.	Stage	Dietary intake per period.			Iron administered.		M.A.	Ca		P		Fe
		Ca.	P	Fe	Clinical dose per day.	Mg. per period.	F	F	U	F	U	F
13	1	6·9	7·3	36			15·5	3·09	0·85	1·37	3·70	37
	2	,,	,,	,,	Reduced iron 135 grains	7179	28·2	3·30	1·03	1·88	3·77	6453
	3	,,	,,	,,			15·3	2·82	0·92	1·30	3·90	503
16	1	2·13	1·32	18·6			8·9	1·83	0·34	1·01	0·93	14
	2	,,	,,	,,	Iron and ammonium citrate 120 grains	5375	20·9	2·13	0·31	1·30	1·00	4785
	3	,,	,,	,,			8·9	1·95	0·28	1·08	0·97	76
18	1	6·0	5·9	72			19·1	4·20	0·69	2·50	2·67	. 34
	2	,,	,,	,,	Iron and ammonium citrate 80 grains	6000	30·7	4·94	0·63	3·02	2·16	5555
	3	,,	,,	,,			18·5	4·89	0·55	2·03	2·58	195
20	1	4·4	5·6	35			9·8	2·31	0·53	0·93	2·78	23
	2	,,	,,	,,	Blaud's pill 120 grains	2752	18·0	2·42	0·49	1·34	1·99	1556
	3	,,	,,	,,			14·9	2·76	0·69	1·47	2·78	582
21	1	3·60	4·14	35			13·5	2·20	0·62	0·72	2·93	28
	2	,,	,,	,,	Iron and ammonium citrate 90 grains	3666	20·6	2·38	0·59	1·92	1·63	3140
	3	,,	,,	,,			8·5	2·09	0·51	0·83	3·10	45

Calcium and phosphorus figures in grams.   Iron figures in milligrams.

M.A. = grams of mineral ash in fæces ;   F = fæces ;   U = urine.

that this point could not be settled.   For what it is worth, however, analysis of a few of the stools showed very much greater sulphur content in the iron stage than in the pre-iron stage.

It is of practical importance to know if the observed increase (averaging 39%) in fæcal phosphorus as a result of iron therapy is sufficient to cause a negative phosphorus balance.   The phosphorus balance is shown in Fig. 3. In Case 13 the small increase in fæcal phosphorus merely decreases a large positive phosphorus balance.   In Cases 18, 20 and 21 the increase in fæcal phosphorus is compensated for by a decrease in urinary phosphorus.   It may seem strange that these patients had such large positive phosphorus balances, but probably the reason for this is that they come from homes in which they had lived on poor or inadequate diets and were placed in hospital on diets which were high in phosphorus.
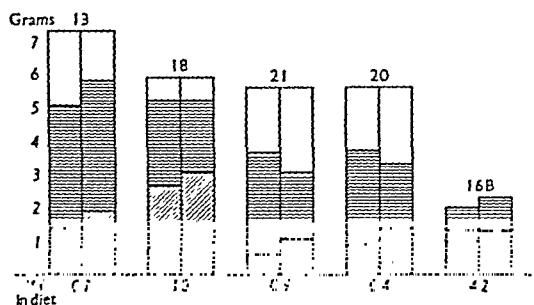
Fig. 3. The effect of iron on the exchange of phosphourus.   Five cases individually.   In each case the two columns represent average weights per period of the first and second stages. Diagonal hatching represents phosphorus in the stools, and transverse hatching phosphorus in the urine.   The dotted line represents the intake of phosphorus.

In Case 16B, however, the diet was markedly deficient in phosphorus so that the patient was in negative balance in the control period.   In this case the administration of iron caused the negative phosphorus balance to become more markedly negative.

DISCUSSION.

It was pointed out in the first paragraph of this paper that the administration of large doses of iron to rats led to phosphorus deficiency rickets when the ratio of iron to phosphorus in the food was a high as 1·5. The addition of extra phosphorus to the diet sufficient to reduce the ratio to 0·8 always prevented rickets.   In the five human cases studied by balance experiments it is apparent that iron therapy does increase the fæcal phosphorus output, and it is presumed that, as in the case of the rats, the mechanism at work is a precipitation of phosphorus in the intestine by the iron. Provided, however, that the phosphorus content of the diet is adequate, there is no danger of producing a state of phosphorus deficiency.   In four cases with ratios of iron to phosphorus up to 1·0 in the intake the effect on the phosphorus balance was negligible.   It is obvious, however, from Case

16B that there must be a critical level of phosphorus intake below which iron therapy can convert a positive phosphorus balance into a negative one.

Large doses of iron have, however, so thoroughly proved their value in clinical medicine that there is no reason to abandon them. It may be possible to show in the future what constitutes the unexplained portion of the rise in fæcal mineral ash resulting from iron therapy, but in the meantime large doses of iron should undoubtedly be used, wherever they are needed, provided care is taken to see that the intake of phosphorus is optimal.*

SUMMARY.

1. The results of calcium, phosphorus, and iron balance studies on five patients treated with large doses of iron are presented.

2. In the five cases there was as a result of iron therapy an average increase of 29% in the fæcal mineral ash, exluding the weight of unabsorbed iron.

3. Part of this increase was due to an average increase of 39% in the fæcal phosphorus.

4. It is suggested that an increase of fæcal sulphur may have been responsible for another part.

5. Fæcal calcium was not increased.

6. The mechanism of the increase in fæcal phosphorus is believed to be the same as that which causes iron rickets in rats, namely a precipitation of phosphorus as insoluble ferric phosphate in the intestine. It is believed, however, that there is very little risk of producing phosphorus deficiency in man, provided the phosphorus content of the diet is adequate.

CLINICAL DETAILS OF CASES.

Case 13. Male, aged 21 years. A case of disseminated sclerosis of three years duration. Blood count : r.b.c., 5,130,000. Hb. 98%. w.b.c., 8,220.

Case 16B. Female, aged 14 years. Convalescent from acute rheumatic fever. The patient had had a hæmoglobin of 67% 3 months previously and had had iron therapy for six weeks about a month before the experiment. At the time of the experiment her blood count was : r.b.c., 5,000,000. Hb. 86%. w.b.c., 6,400.

Case 18. Male, aged 7 years. Convalescent from pleurisy with effusion. Blood count : r.b.c., 5,000,000. Hb. 87%. w.b.c., 7,200.

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* The fate of the administered iron as determined by balance experiments is discussed elsewhere (Brock and Hunter, 1937).

Case 20. Female, aged 30, single. A case of idiopathic hypochromic anæmia. Bluish sclerotics. Normal tongue and finger nails.

Satisfactory dietary history and no record of excessive loss of blood :

Blood count : r.b.c., 3,460,000. Hb. 45%. C.I., 0·68. w.b.c., 3,800.

Test meal : No free HCl in first 1½ hours. After histamine, free HCl 0·25%.

Satisfactory response to iron therapy.

Case 21. Female, aged 38, single. A case of idiopathic hypochromic anæmia. Dietary history satisfactory. No excessive blood loss. Dysphagia and brittle finger nails. Slight marginal atrophy of tongue.

Blood count : r.b.c., 3,600,000. Hb., 45%.

Test meal : No free HCl in first 1½ hours. After histamine, 0·15% free HCl. Satisfactory response to iron therapy.

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OBSERVATIONS ON A CASE OF FAMILIAL PERIODIC PARALYSIS.* -

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ALTHOUGH familial periodic paralysis has been known and studied for over fifty years, the cause of the disease and the mechanism whereby the paralytic attacks are produced have remained obscure. The patient on whom the observations here recorded were made showed nearly all of the characteristic features of the condition.

Case record.

S.P., male, was born in London, in 1914, of English parentage. He says that his father suffered from periodic attacks of paralysis from the age of 14 until his death of pneumonia at 63, the attacks becoming less severe in later life ; and that his father's father had similar attacks. His mother can give only vague confirmation of this statement. No other children of his father are known. His childhood was healthy ; he was normally intelligent at school, but a little clumsy at games. From the age of $14\frac{1}{2}$ onwards he experienced attacks of paralysis at irregular intervals of several weeks. For some years he was regarded either as a hysteric or as a malingerer, by his mother and his step-father, by doctors who saw him, and by those under whose charge he was placed in a reformatory school; he was treated, he says, with considerable severity, and has since been erratic in his general behaviour, subject to periods of mental depression, and inclined to act impulsively. At Guy's Hospital in 1935 his condition was recognised as familial periodic paralysis and he has frequented various hospitals in and near London since then.

The figures are all within normal limits except the serum potassium value during an attack, which is low (approximate normal range is 16-20 mg. per 100 c.c.). In another similar attack the serum potassium value was found to be 12·6 mg. per 100 c.c. compared with 17·8 mg. per 100 c.c. in an interval.*

The patient then insisted on leaving the hospital, but was re-admitted in April-May, 1936, when he experienced an unusually severe spontaneous attack, lasting about 53 hours. Blood examinations gave the results shown in Table II.

The patient next came under observation (M.W.) in St. Leonard's Hospital in June, 1936, when a spontaneous attack was observed. Since an abnormally low serum potassium level had been found during attacks
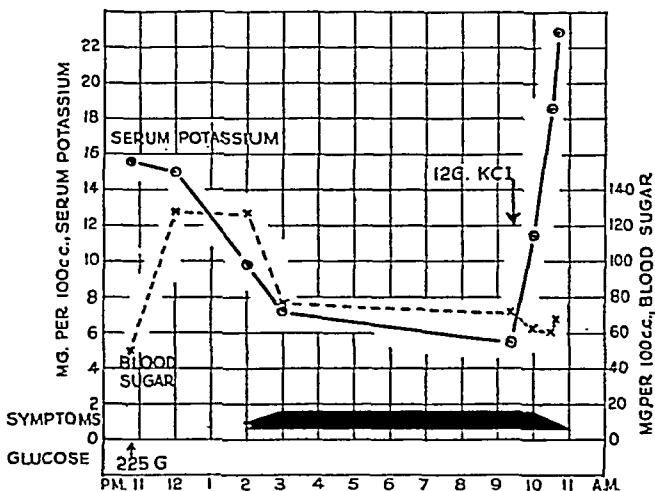
Fig. 1. Blood sugar and serum potassium during an attack of paralysis induced by orally administered glucose.

on three occasions, administration of potassium by mouth was tried. 12 g. of potassium chloride were given in water, when the attack had lasted about 16 hours. The affected limbs began to regain their power in 10 minutes and in an hour had returned to their normal state. A similar effect was obtained in an induced attack, the details of which, with blood sugar and serum potassium curves, are given in Fig. 1. It was also shown that both lævulose and carbohydrate in the form of water biscuits are as effective as glucose in inducing attacks.

In August, 1936, the patient was admitted to Hammersmith Hospital under the care of two of us (R.S.A. and L.I.M.C.), and further investigation became possible. The induction of an attack by glucose was repeated (Fig. 2), and the attack was terminated within an hour or two of its onset by 12 g. of potassium chloride given orally. The values plotted in Fig. 2

* These preliminary findings were recorded in a letter to *The Lancet*, 1935, 2, 47.

for serum calcium and serum magnesium show again that these ions undergo no change in concentration at all comparable with the potassium changes.

Although glucose administration induces attacks, the blood sugar levels recorded above are not abnormal. The only abnormality that we have detected in carbohydrate metabolism is a tendency for the fasting blood sugar level to be low and a slightly increased " tolerance " for glucose as shown by this test : blood sugar fasting, 66 mg. per 100 c.c. ; at half-hour intervals after 50 g. glucose by mouth, 84, 101, 80, 73 mg. per 100 c.c.. Sugar was absent from the urine during the test, and has not been found in any of the numerous specimens examined at other times. It was clear that the rise in blood sugar level which occurs after 250 g. glucose is given by mouth does not by itself cause paralysis, but it was conceivable that it
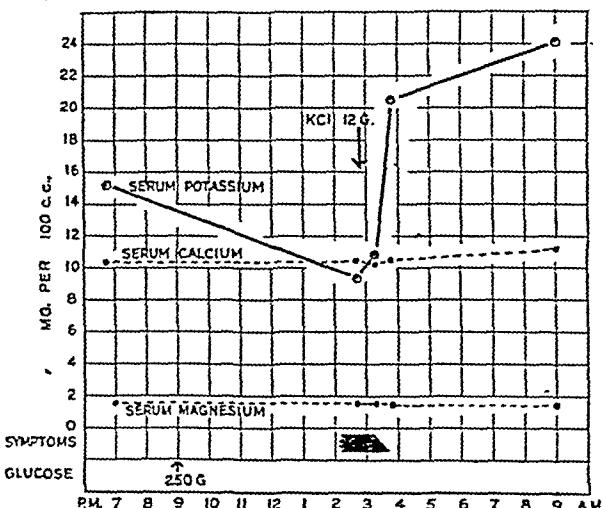
Fig. 2.   Serum potassium calcium and magnesium during an attack of paralysis induced by orally administered glucose.

might, in trigger fashion, start a sequence of events leading eventually to paralysis. To test this possibility 40 g. of glucose in 20% solution were given intravenously at 9 p.m. at a rate judged sufficient to raise the blood sugar level to 200 to 300 mg. per 100 c.c.. The patient slept from 10 p.m. till 12.30 a.m.. No weakness or paralysis developed.

The alternative hypothesis, that post-hyperglycæmic hypoglycæmia might be responsible not directly but indirectly for the development of paralysis, was suggested by the fact that the glucose-induced attacks did not usually manifest themselves until about 5 hours after the glucose had been given. To test this, 20 units of insulin were given subcutaneously at 11 p.m. after a day of normal meals. The patient slept, but woke at midnight with mild hypoglycæmic symptoms, including hunger ; a nurse gave him water and bread and butter, and he slept again, to wake at 4.30

a.m. with paralysed limbs.   Details of this experiment are given in Fig. 3, which shows the typical fall in serum potassium level, and the usual rapid return to normal accompanying the abolition of the paralysis by potassium chloride.   The blood sugar level was no doubt low at midnight, but had returned to normal, by the time the patient woke paralysed.
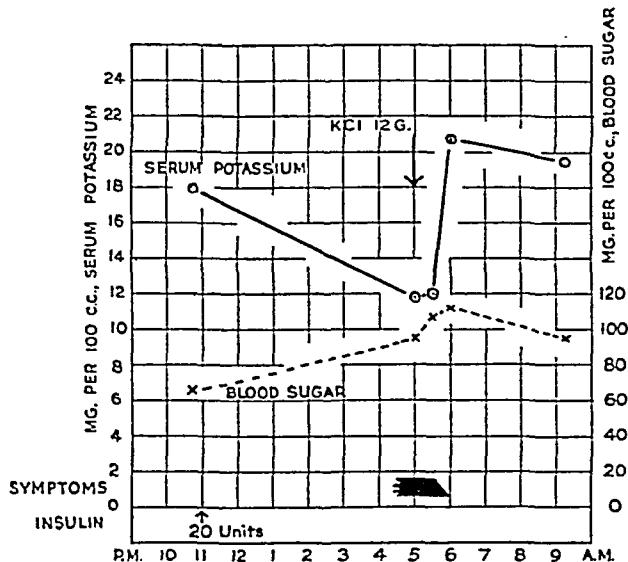
Fig. 3.   Blood sugar and serum potassium in attack induced at night by insulin.
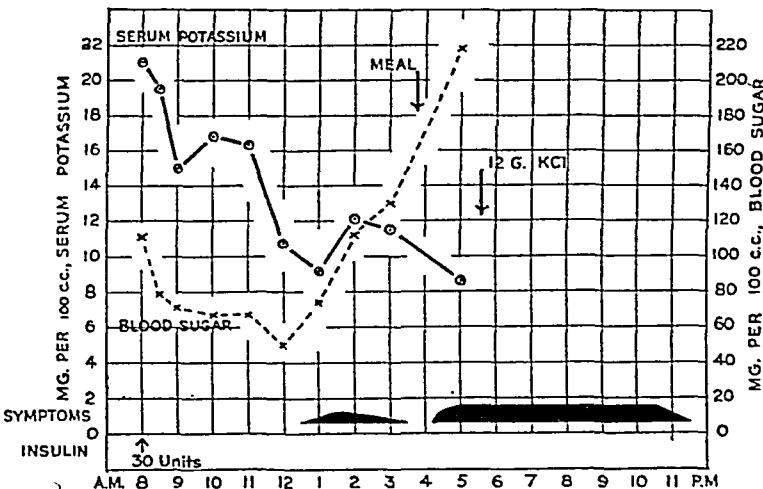
Fig. 4.   Blood sugar and serum potassium in an attack induced by insulin in the day-time.

Fig. 4 gives the result of a similar experiment carried out in the daytime, the patient lying quietly in bed and having no food except half a pint of milk at 6 a.m.   Following 30 units of insulin at 8 a.m. the serum potassium level fell progressively and when, between midday and 1 p.m., it reached about 10 mg. per 100 c.c. an incomplete attack of paralysis developed.   The

potassium level then rose slightly and the symptoms passed off spontaneously about 3 p.m.. At 3.30 p.m. the patient rose, washed, dressed, and ate a generous tea. While he sat at table his legs became weak and he was carried back to bed. A full attack of paralysis rapidly developed, and persisted, in spite of the administration of potassium chloride, until 11.30 p.m.. This is one of the only two occasions on which potassium chloride has failed to abolish the paralysis within an hour. In a score or more of other attacks, spontaneous or induced, it has produced a typical recovery. The two exceptions cannot at present be explained ; serum potassium figures during the prolonged paralysis are not available in either case..

In two further experiments (Figs. 5 and 6) glucose by mouth and insulin subcutaneously were given simultaneously in repeated doses. The experiments were made in the daytime, the patient having no food except half-a-pint of milk at 6 a.m., and lying in bed throughout. In each case a marked fall of serum potassium level occurred Weakness of the limbs appeared

TABLE III.

Blood sugar and serum potassium values during 24 hours without paralysis.

	Serum potassium mg. per 100 c.c.	Blood sugar mg. per 100 c.c.
12 noon	17·6	86
3 p.m.	17·2	92
6 p.m.	16·3	87
9 p.m.	17·9	113
12 midnight	15·8	84
3 a.m.	18·1	86
6 a.m.	16·4	66
9 a.m.	17·3	66

when the potassium figure was about 12 mg. per 100 c.c. and increased to full paralysis, the increase being more rapid in the case (Fig. 6) where the serum potassium fall was more steep. Prostigmin given subcutaneously in doses of 1·5 mg. and 2·5 mg., with atropine, had no effect on the paralysis.

At this stage certain control observations are relevant. The patient's serum potassium values, apart from attacks, lie within the normal range (16 to 20 mg. per 100 c.c.). Thus the initial readings in 16 experiments have lain between 15·2 and 20·7, average 18·75 mg. per 100 c.c., and values found at three-hourly intervals during a day when he had normal meals and sleep are shown in Table III.

The behaviour of serum potassium has been observed in other subjects following the administration of (1) glucose, (2) insulin, (3) glucose plus insulin, in conditions similar to those of the experiments on the patient. Typical results are shown in Figs. 7 and 8. In each case a fall in serum potassium
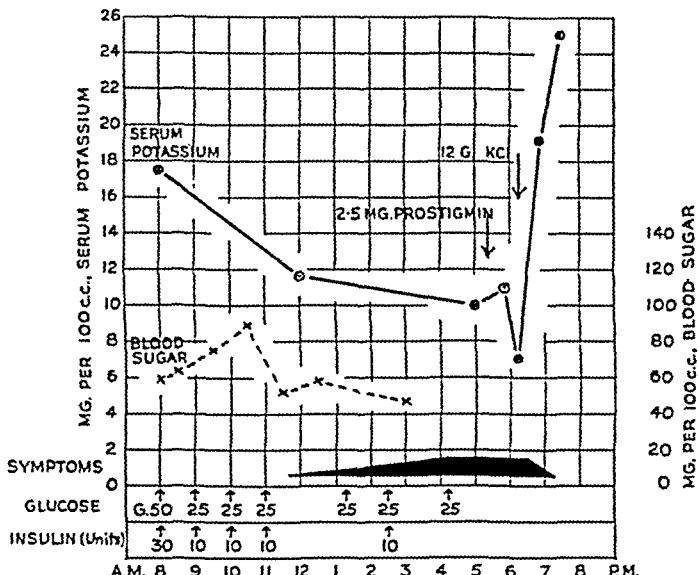

Fig. 5. Blood sugar and serum potassium in an attack induced by glucose and insulin (1 mg adrenalin hydrochloride was given intramuscularly at 5.53 p.m.).
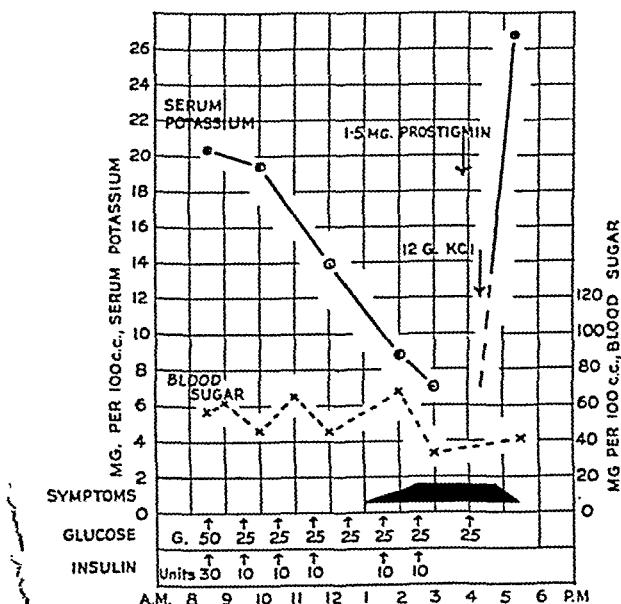

Fig. 6. Blood sugar and serum potassium in an attack induced by glucose and insulin.

level occurred, but the fall was less marked and less prolonged than those occurring in the patient and values lower than 15·7 mg. per 100 c.c. were not recorded.

Discussion.

We have found only one observation in past records suggesting a relation between serum potassium level and the development of muscular weakness in familial periodic paralysis.* Biemond and Daniels (1) recorded a value of 13·38 mg. per 100 c.c. in a mild spontaneous attack compared with 17·87 mg. per 100 c.c. in an interval. They did not, however, appreciate its significance. The observations described above establish the relationship in this patient as a close and consistent one. When the serum potassium level falls to about 12 mg. per 100 c.c. the muscular weakness begins to develop, and levels of 10 mg. per 100 c.c. or lower are associated with full paralysis. When the level rises again after potassium chloride has been given by mouth the muscular power returns *pari passu.* The neuro-muscular
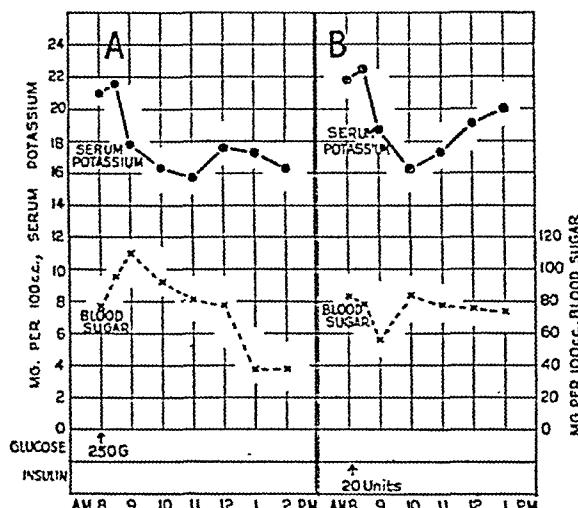
Fig. 7. A. Blood sugar and serum potassium in a male subject aged 27 following the ingestion of 250 g. glucose.

B. Blood sugar and serum potassium in a male subject aged 31 following the injection of 20 units of insulin.

disturbance has been shown by electrical stimulation to lie peripherally. It must be supposed, therefore, that a lowered potassium concentration in blood and tissue fluids interferes with one or more of the following three processes :—(a) the passage of the nerve impulse along the fibre of the nerve concerned, (b) its transmission across the neuromuscular junction, (c) the response of the muscle fibre to the impulse reaching its motor end plate. We have no indication as to which of these processes is interfered with, except the negative inference from the prostigmin observations that the interference is not of the kind that occurs in myasthenia gravis.

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* M. S. Herrington (*Jour. Amer. Med. Ass.* 1937, **108**, 1339) has recently described the successful treatment of incipient attacks, in two patients, over a period of 2 years, by potassium citrate in 5 g. doses.

Further, it is not clear if a low serum potassium level is the only factor required to produce paralysis. The neuro-muscular apparatus may itself be abnormal, as is suggested by Biemond and Daniels' observation that some subjects of periodic paralysis developed in later life an atrophy of those muscles that had been most affected by paralysis. A neuro-muscular abnormality would provide an easy explanation for the fact that paralysis occurs in limb and trunk muscles only, sparing those of the head and face, but normal variations in the sensitivity of different muscles to low potassium concentrations can equally well be postulated. Moreover, there are a few records of serum potassium levels in man lower than 10 mg. per 100 c.c., following the injection of insulin, and no mention of paralysis accompanies them. Harrop and Benedict (3) found values of 9·4 and 8·5 mg. per 100 c.c. in two diabetic patients, and Kerr (4) found 7·2 mg. per 100 c.c. in a diabetic, in both cases after insulin.
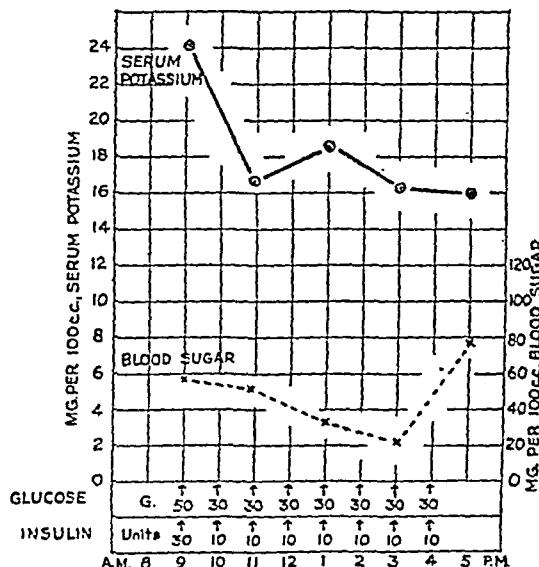
Fig. 8. Blood sugar and serum potassium in a male subject aged 45 during repeated administration of glucose and insulin.

We have failed hitherto to reduce the serum potassium level in subjects other than the patient to 12 mg. per 100 c.c. or lower. The presence or absence of a neuro-muscular abnormality in the patient therefore remains undecided. On the other hand, a " metabolic " abnormality is strongly suggested, namely, an undue lability of the serum potassium level. The fact that the action of injected insulin is accompanied by a fall in serum potassium level in man and animals has been known for many years (Harrop and Benedict (3); Briggs, Koechig, Doisy and Weber (2); Kerr (4)). We have shown that a similar fall occurs after ingestion of glucose, with or without injection of insulin, and that it can occur independently of hypoglycæmia (*see* Fig. 7A). This makes it unlikely that the fall is part of a compensatory

physiological response to hypoglycæmia. The phenomenon is therefore better described as a fall in serum potassium level accompanying the passage of sugar out of the blood into the tissues. It is a normal phenomenon, but in this patient subject to periodic paralysis it appears to be exaggerated. That is his metabolic abnormality, so far as it can be defined at present.

SUMMARY.

In a patient suffering from familial periodic paralysis the serum potassium level has been found abnormally low during paralytic attacks. The serum potassium level can be lowered to an abnormal extent by the administration of large amounts of glucose by mouth, by the injection of insulin, and especially by combined injection of insulin and ingestion of glucose. When it falls below 12 mg. per 100 c.c. paralysis develops. Administration of potassium chloride by mouth raises the serum potassium level and abolishes the paralysis. It is concluded that the fall in serum potassium concentration, normally associated with the passage of glucose from blood into tissues, is in this patient abnormally great, and that lowering of potassium concentration either blocks neuro-muscular transmission or inhibits the contractile response in the muscles affected.

APPENDIX.

The chemical estimations, with the exception of the blood sugar values recorded in Figs. 2-8 and Table 3, were made by E.N.A.. The following methods were used :—

K : Cobaltinitrite precipitation, direct on serum (Kramer and Tisdall, J. Biol. Chem., 1921, 46, 339).
In all cases the serum was separated within an hour after withdrawal of the blood to avoid any risk of potassium loss from the corpuscles. (*See* Watchorn and McCance, Biochem. Journ., 1933, 27, 1107).

Na : Uranyl gravimetric method (Butler and Tuthill, J. Biol. Chem., 1931, 93, 171).

Ca : Precipitated as oxalate, and washed by Clark and Collip's method, (J. Biol. Chem., 1925, 63, 461).

Mg : Colorimetric magnesium ammonium phosphate precipitate. (P determined by Kuttner and Lichtenstein method).

Cl : Open Carius method (Van Slyke and Sendroy, J. Biol. Chem., 1925, 58, 523).

P : Colorimetric using stannous chloride as reducing agent (Kuttner and Lichtenstein, J. Biol. Chem., 1930, 86, 671).

HCO_3 : CO_2 capacity method, using Van Slyke volumetric apparatus (Van Slyke and Cullen, J. Biol. Chem., 1917, 30, 289).

Protein : Microkjeldahl (Howe, J. Biol. Chem., 1921, 49, 109).

Urea : Colorimetric micro-urease (Archer and Robb, Quart. J. Med., 1925, 18, 274).

Sugar : Schaffer-Hartmann (Harding's modification of the Schaffer-Hartmann method—Harding and Downs, J. Biol. Chem., 1933, 101, 487), or Hagedorn-Jensen method (Biochem. Z., 1923, 135, 46), using unlaked capillary blood.

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THE EFFECT OF ASPHYXIA AND COCAINE ON NERVES BELONGING TO THE NOCIFENSOR SYSTEM.

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It has been recorded in a recent paper (4) that when a small fold of human skin is crushed in the tip of forceps, this injury may give rise to widespread hyperalgesia around the crush. The hyperalgesia is produced by the local action of a system of nerves, which, so it is concluded, belong to a hitherto undescribed system, being neither sympathetic nor sensory, and for which the name " nocifensor system " has been suggested. The present observations attempt to explore the characteristics of these nerves further by testing the effects of asphyxia upon them.

Asphyxia and cutaneous nerve fibres.

Six years ago, in conjunction with Drs. Pickering and Rothschild (5) I recorded the march of events in the human arm, to which the circulation had been stopped. We showed that in this ischæmia nerve fibres supplying voluntary and involuntary muscle, and various kinds of sensation, are affected after different intervals of time. Although we put forward proof, so I think, that this interesting and useful differentiation is the result of asphyxia, Bishop and his collaborators (1) were inclined subsequently to attribute the effects to direct pressure on the nerve trunks. But their view has been abandoned since, and our conclusion that asphyxia is responsible seems now to be accepted. Gasser and his collaborators (2, 3), having confirmed the relevant observations on man, have adopted our method to differentiate between different groups of fibres (classing them, however, according to their conduction rates rather than according to their functions) in the nerve trunks of animals.

The facts that we elicited and that are relevant at present are as follows. If the arm is kept at body temperature and the circulation is stopped by pneumatic pressure upon the upper arm, sensation is first affected at the

tips of the fingers in about 13 to 15 minutes. Numbness is noticed and this numbness spreads gradually up the fingers, hand, and arm until it reaches the upper arm in about 30 minutes. Impairment is ultimately followed by loss of sense of touch, the latter following the former at a given level after an interval of about 5 minutes. It is clear that sense of cold, warmth, and pain are lost later than the sense of touch, and the order of loss is probably, touch, cold, and warmth and pain ; loss of pilomotor response, the only form of sympathetic nerve response so far tested, is long delayed.

A simple but precise statement as to the order in which the responses are affected is rendered difficult by the gradualness of the changes. When the hand has lost all sense of touch, it has also lost sense of deep pressure, so that coarse contacts remain unrecognised. Though the hand in this state is still capable of clearly appreciating cold, warmth, and pain, none of these three is at this time unaffected ; the loss of each of the three sensations is gradual, just as it is gradual in the case of touch. Thus, to take pain as an example, the immediate response to light pricks is not recognised so often at the stage when the skin of the hand has become anæsthetic as when it is normal, though a sharp stab with a needle is felt at once and invariably. Regarding the first changes to light pricks as significant, then it may be said that pain sense is first recognised to become affected at or about the time when the skin loses all sense of touch ; it may be a little earlier than this, but it is never nearly so early in onset as numbness. Painful response to needle pricks is not lost altogether until very much later. Very similar statements may be made of cold and warmth ; both these senses fail gradually but both persist in some degree after sense of touch is lost.

Asphyxia and the hyperalgesia reaction.

Our first object has been to try and determine where in this range of cutaneous nerve change the hyperalgesia reaction displayed through nocifensor nerves falls out. Subjects have been chosen who give conspicuous responses to small crushes of the skin. The arm is rendered ischæmic in the usual way by pneumatic compression of the upper arm, the forearm and hand lying immersed in water at 35°C.. The march of touch and pain loss is closely watched, and a little fold of the skin, about 5 to 10 cm. above the wrist and on the ventral surface of the forearm, is crushed at various times relative to the other events in separate tests. After the skin is crushed, about 5 minutes are allowed for the products of the crush to act through the nocifensor nerves ; the reaction is then stopped by infiltrating the crushed area with 1% novocaine intradermally. The circulation is then released and, after subsequent recovery from the sensory impairment due to ischæmia of the skin, and from the reactive hyperæmia, the skin is tested for hyperalgesia around the crushed and still anæsthetic skin.

The results may be summarised. If the skin is crushed after sense of touch in it is lost, or when it is numb and shortly before it loses touch sense, hyperalgesia fails to appear around the crush, either at the time, or after

the circulation is released. If the skin is crushed when numbness in it is just detectible, or a minute or two before it becomes numb, then hyperalgesia develops subsequently, but it is not developed fully as compared with simultaneous controls. If the skin is crushed before numbness reaches it, and remains without numbness for most of the 5 min. period, hyperalgesia develops fully.

It is noted that if in a control the skin is crushed first and the arm is then asphyxiated, the hyperalgesia around the crush often continues to be recognisable until after the skin around it has completely lost sense of touch. It is also to be noted that hyperalgesia is found in all acceptable tests around the crush, once the crushed skin has recovered from local anæsthesia.

Thus, if we assume that asphyxia interferes with the production of hyperalgesia by paralysing conduction in nocifensor nerves, these nerves must be thrown out of action very early; and the earliness is emphasised by a further consideration. The times at which the nocifensor nerves are thrown out of action may be accepted on this basis without correction, for they are tested at the place of the reaction, which occurs in an axonic system that must be in the main quite local. But the asphyxial test acts differently in the case of the sensory nerves. Numbness, when it first appears in the skin, is produced by asphyxia of fibres lying directly beneath the pneumatic cuff; the same nerves in their course through the lower parts of the arm are unaffected until later times. This was shown in our earlier work by placing a second pneumatic cuff below the first and releasing the former; this procedure always causes the upper borders of numbness and anæsthesia to recede. Experiments of this kind show that, when the cuff is above the elbow, the function of those of the underlying sensory nerve fibres which supply the wrist is lost about 10 minutes before the function of the same fibres is lost through asphyxia in the region of the wrist itself. Making due allowance for this delay, it would seem likely that the local nocifensor nerves are the first to be thrown out of action, being thrown out before the touch nerves and long before those of cold, warmth, and pain.

Block by band of asphyxia. The observations on asphyxia just described are not, however, conclusive. It is an assumption that asphyxia prevents hyperalgesia developing by acting on the nocifensor nerves; it is possible that it acts by interfering in some way with the effector mechanism of these nerves. To test this possibility another method has been used. A rubber band 12 mm. in width, is stretched around the forearm and fastened in place at a tension sufficient to stop the circulation to the skin underlying it. A pressure of 60 mm. Hg. has been shown to suffice for this when blood pressure is normal. The rubber band, reduced to 8 or 9 mm. width when stretched and in place, should be sufficiently tight to depress the skin without throwing it into folds; a convenient tension is easily judged with practice. When removed after many minutes, the skin presents a flat depression, which at once flushes bright red. This reactive hyperæmia, which appears at release, indicates that the circulation has been arrested previously. To prevent

congestion of the arm, a saddle shaped bridge is placed on the dorsal surface of the arm over a main subcutaneous vein, and the rubber band runs over this bridge, which guards the vein from the pressure.   The front of the forearm is used for the experiment.

The band is left in place for varying times and a little fold of skin is then crushed immediately distal to the band.   Just before the crush, the skin above and below the band is closely examined to ensure that any hyperalgesia subsequently appearing may be recognised with certainty.   If the band has been in place for 20 to 25 minutes, hyperalgesia develops above and below it.   If it has been in place for 25 to 30 minutes, hyperalgesia develops up to the band on the side of the crush but not beyond it.   If the band is then removed, hyperalgesia soon appears in the skin proximal to the place where
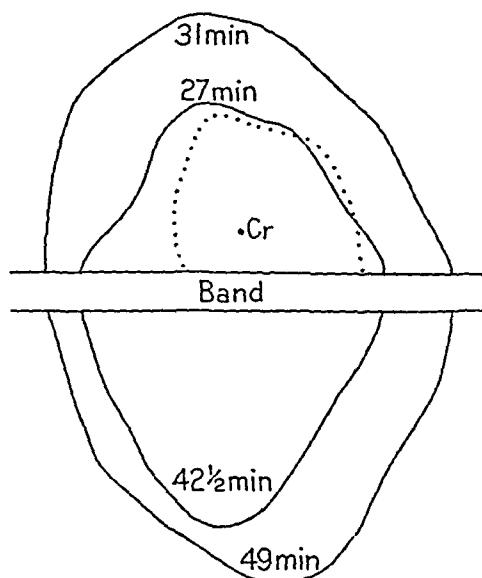
Fig. 1. H. (× ¼).   The top of the diagram is distal. The rubber band was stretched over the skin of the arm. 25 min. later the skin was crushed at *Cr.*. At 27 min. an area of hyperalgesia had appeared as shown around the crush below the band ; this area became extended by the 31st min.. At this time too a brilliant flare was present on the skin below the band (dotted outline). Above the band there was no hyperalgesia and only one small area where a little flare developed. At 40 min. the rubber band was removed. The hyperalgesia above the band developed subsequently and was outlined at 42¼ and 49 min..

the band lay.   The development of hyperalgesia above a band that has been in place for 20 minutes is of full extent and intensity, and takes place hand in hand with that of the hyperalgesia below the band.   When the band has been in place for 25 min. the hyperalgesia may be delayed in its appearance above the band and may remain under-developed in intensity and extent until after the band is removed.

These observations show that asphyxia of the skin of about 25 min. duration will block the passage of impulses through the nocifensor nerves

of the skin ; they clearly indicate that the action is on the nerves and not on the effector mechanism, when the whole arm is asphyxiated. This method also allows a direct measure of the state of skin to be taken. Controls show that skin deprived of its blood supply by direct pressure for about 25 min. presents very slight numbness to gentle touches ; it is far from being anæsthetic ; it presents no change in its response to needle pricks.

The observations with the band, and those in which the whole arm is asphyxiated, are in good accord.

Cocaine and the hyperalgesia reaction.

It is known that the order in which cutaneous nerve fibres, serving sensory and other functions, are paralysed by cocaine is in general the reverse of that in which they are paralysed by asphyxia (*see* 2 and 3). Thus pain fibres are paralysed before touch fibres. The nerves underlying the hyperalgesia reaction appear to be paralysed very early by asphyxia ; it would be consistent for them to be paralysed very late by cocaine.

Cocaine has been driven into the skin electrophoretically, using a 1% solution, and a current of 80 to 150 or 200 microamp. per sq. cm.. The weaker current, passed for 5 min., usually makes the skin analgesic to needle prick and distinctly dulls but does not abolish sense of touch. The stronger current passed for 9 to 12 min. usually abolishes all sense of touch. A circular area of the forearm of 2 cm. diameter, is treated in this way, the bloodflow to the arm is then arrested to prevent recovery, and the condition of sensation in the skin is quickly examined. The state being ascertained, a little fold of skin is crushed within the analgesic zone ; the crush should be quite unfelt. If the electrophoresis has abolished all responses to needle prick and has so far depressed that it has almost abolished the sense of touch, the usual reaction to the crush is unaffected ; that is to say, hyperalgesia develops in a wide area of surrounding skin in the usual time, and this area of hyperalgesia is of full extent and intensity, showing no increase after the circulation is released and the cocainised skin recovers. This result is invariable. If, before the skin is crushed, the cocainisation has been pressed farther, so that touches are only felt if the skin surface is definitely deformed by them and so that pain sense is lost, not only to needle pricks but to the more searching test of applying hot metal to the skin (copper at 63°C. for 2/5ths or 3/5ths of a second) then the result is the same, or there may be a partial reaction, or hyperalgesia may fail to appear until after the anæsthetic skin recovers consequent upon the release of the circulation. Thus, the reaction is not blocked with certainty in skin until or after a grade of cocainisation has been reached that renders the skin both analgesic and anæsthetic in the full sense. Although complete paralysis of the deepest sensory nerve fibres of the skin may still be questionable, these cannot be unaffected ; it is however most improbable that with this grade of affection of the sensory nerves as a whole, the hyperalgesia reaction would be unaffected in rate of development and extent if provoked through them.

These observations on cocaine, and those already described on asphyxia are in physiological harmony; they support the conclusion reached in a previous paper, that the nocifensor nerves, though travelling through the posterior roots, are not sensory but belong to a separate system.

Asphyxia and the nerves governing the flare.

In my earlier paper I suggested that the area of hyperalgesia and the area of vascular flare, which soon appears around small injuries of the skin, may possibly be provoked through one and the same system, namely the nocifensor system of nerves. The present observations enable the effects of asphyxia on the nerves underlying hyperalgesia and flare to be compared. The tests are carried out in the same way as those previously described; the circulation to the arm is arrested, the skin of the lower forearm is crushed with forceps and after 5 min. is anæsthetised locally and the circulation released. In the case of the flare it is necessary to use a control, so that two flares may be compared for size and brightness. This can be done by crushing the skin of the forearm at two levels, the skin when crushed presenting different states of sensory defect; but this method has the disadvantage that difference of level is apt to affect the extent and brightness of the flare. Another method is to crush the skin at the same level in two places on the arm, but at different times; though here there is the disadvantage that the two areas of hyperalgesia are apt to run together and to become confused, so that observations upon hyperalgesia and upon flare are not readily undertaken simultaneously. The third and most satisfactory plan is to use the two arms simultaneously,* arresting the circulation 10 minutes earlier in one than in the other, crushing the skin on both sides simultaneously, anæsthetising each 5 min. later and at once restoring the circulation to both. The flares can thus be compared directly at all stages from the time of their appearance when reactive hyperæmia fades away. The results obtained by all methods are in substantial agreement. The flare is abolished if the skin is very numb or is actually without sense of touch during the greater part of the 5 min. period. The flare is reduced in size and brightness if numbness has ascended the arm to a point well above the crush at the time this is made. Thus the duration of asphyxia required to abolish the " flare " reaction is of the same general order as that required to abolish the hyperalgesia reaction. The flare, however, is affected later than is hyperalgesia.

The band method is not so well adapted to the study of the flare as to that of hyperalgesia, because the area distinctly reddened is relatively small and the band covers much of it. Moreover the flare is known to be conducted partly by nerves lying deep to the skin. Flare is not blocked

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* In such tests, the two arms being treated unequally, 3 or 4 days should be allowed to elapse before the same subject is used again. An arm asphyxiated yesterday is not completely recovered by to-day, the time required to produce a given grade of sensory change is shortened.

as readily as hyperalgesia; it may be blocked by a 30 min. asphyxial band, but much more often it is reduced in intensity and extent.

SUMMARY.

The development of hyperalgesia around a small crush of the skin can be prevented by asphyxiating the skin; and the spread of hyperalgesia can be blocked by asphyxiating a narrow band of skin. The duration of asphyxia required is such as has little appreciable effect on the functions of the sensory nerves of the skin. Asphyxia must be carried a little farther to prevent the development of the " flare " or to interfere with the spread of the " flare " in response to the same injury. Asphyxia stops hyperalgesia from developing by paralysing the nerves concerned ; these nerves are paralysed before the sensory nerves.

Cocaine introduced electrophoretically into the skin paralyses the pain nerves completely and the touch nerves entirely or almost entirely before it interferes with the hyperalgesia reaction.

The conclusion previously formed that the nocifensor nerves, though belonging to the posterior root system, are not sensory but belong to a special system, is supported by both these series of observations.

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THE DOUBLE PAIN RESPONSE OF THE HUMAN SKIN TO A SINGLE STIMULUS.*

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In studying the effects of ischæmia upon the arm, it was found by Lewis, Pickering, and Rothschild (8) that sense of touch is affected before pain, and that, when actual anæsthesia has developed, the pain response to a needle prick, though appreciable, seems to be delayed. This observation was confirmed by Gasser (2 and 5), who suggested that the delay in the pain response may be due to the functional survival of slow conducting fibres.

While recently testing the ischæmic limb for pain sense, and attending to the pain produced by needle pricks on normal and ischæmic fingers, we observed that the pain produced from the normal finger is often twofold, a first pain coming almost at once, and a second pain following after a period of delay ; and it seemed to us, in comparing the normal and ischæmic fingers, that the phenomenon previously described as a delayed response in asphyxia, was more correctly interpreted as the simple loss of the 1st response, the 2nd response being now isolated and therefore more readily discerned. Moreover, having recognised the double response to the single prick, we recollected that such an observation had previously been described and on discovering the source found many observations and reflections already recorded.

A double response to a single stimulus was first described by Rosenbach (9) in 1884 in observations attempting to explain the delay of pain response to stimulation, previously observed in cases of spinal cord disease. Rosenbach used heat as stimulus. A similar reaction to needle prick was described by Gad and Goldscheider in 1898, though these workers emphasised the first response as touch and the second as pain. Gad and Goldscheider (4) explained the double response by supposing that the 1st response travels through the posterior columns and that the 2nd is delayed in the grey matter of the spinal cord. Thunberg (10) a few years later clearly recognised that both responses may be painful, measured their latencies, showed that the double response may be elicited not only by prick but by heat, and added

many interesting observations.   Thunberg disagreed with Gad and Gold-scheider's explanation and suggested that the 1st response is due to stimulation of nerve fibres and the 2nd to stimulation of nerve endings.   The lag of the 2nd response was supposed to be due to a hypothetical process intervening between the stimulus and the ultimate excitation of the nerve ending.   We shall not relate the arguments upon which these explanations were based, or those of Alrutz's criticism (1) of Thunberg, or those of the latter's reply, all of which will be found in Thunberg's main article.   We refrain from doing so because the arguments in favour of the several views seem unsubstantial, and because the hypotheses of delay in cord or delay at the nerve ending are clearly disproved, so we think, by the observations we shall describe.

We agree with Thunberg's description of the response to needle pricks. If a fine needle is used, and skin, say of the back of the finger or of the front of the forearm, is stabbed quickly but lightly, preferably by using needle points set up on bristles bending under ascertained tensions of 2 to 4 gram
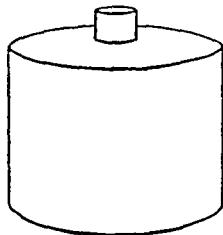
Fig. 1.   (× ⅓.)   Copper cylinder used in stimulating with heat.

after the manner of Frey, the contact gives a simple sense of touch or a quick flash of pain.   After an interval a second pain is felt.   It is important that the skin tested should not be unduly cold, otherwise the 2nd response is unfelt or is not felt clearly.   With light pricks this 2nd response is usually the more intense of the two, the first indeed frequently fails, and the 2nd seems longer lasting and for that reason is more easily described as " stinging"; it seems also to be felt a little more diffusely than the 1st.   Heavier jabs with a needle give two pain responses, the first of which is the more painful and is then apt to attract exclusive attention.

The most certain method of eliciting the double pain response is by using hot metal contacts as the stimulus, as did Thunberg.   We have employed a cylinder of copper, 4 cm. long and 5 cm. diam. ; a mass of metal from which a shorter cylinder of 1 cm. diam. projects (Fig. 1).   The flat surface of the small cylinder is applied to the skin, the large cylinder forms a reservoir of heat for successive tests.   The metal is heated, usually to 63°C., in a water bath, and the contact with the skin lasts 1/5 to 1/2 sec..   Such a contact is usually long enough to be accompanied by a distinct but brief stinging pain, followed after an interval, during which the metal has been removed, by a 2nd sting, which, if the contact has been short, exceeds the 1st sting in its intensity.

Evidence for two sets of pain nerves.

Dissociation by ischœmia. If the circulation to the arm is arrested and the arm is kept in a bath at 34°C. the fingers begin to be numb at their tips in about 15 min. and anæsthetic in about 20 min.. This diminution and loss of touch sense spreads up the limb. At about the time when the fingers become actually anæsthetic (25 to 30 min.), or a little earlier, the 1st response which they give to light needle pricks is lost, the 2nd response being at first undiminished in intensity, but at a later stage reduced. We call it the 2nd response because, when compared with the 2nd response from control skin it comes at the corresponding time (*see* Table I). If in the late stage the finger is jabbed harder with the needle, the 1st response is felt also.

TABLE I.

Stimulus to 2nd response in sec..

Series 1. Control.				Series 2. Asphyxia.					
E.E.P.	1·1	1·1	1·1	1·0	1·2	1·2	1·1	1·2	
	1·0	1·0	1·1		1·2	1·1			
		av. = 1·1				av. = 1·2			
T.L.	1·5	1·4	1·4	1·4	1·2	1·3	1·1	1·2	1·2	1·2
		av. = 1·4				av. = 1·2				

The method of measuring the intervals is described on page 71.

The asphyxia must be carried on for longer periods before the 1st response to heavy stimulation is lost (50 to 75 mins), and for still longer periods before both responses are lost (75 to 100 mins). It is undesirable to arrest the circulation to a limb repeatedly for periods much longer than a half hour; so in long asphyxiation we first bind off the finger at its base, remove the bandage after 75 min. have elapsed, but arrest the bloodflow to the arm immediately before doing so and continue the asphyxia for a further period of half an hour. Thus the arm is safeguarded from long asphyxia and the finger has been freed from the bandage long before tests begin. Using the heat stimulus, corresponding results are obtained, the 1st response declines and vanishes, before the 2nd is noticeably affected.

In an earlier paper from this laboratory a special pressure clamp was described, which allows a short stretch of the nerves of the upper arm to be rendered ischæmic while bloodflow to the whole of the lower arm is fully maintained. We have used this clamp to ascertain if the dissociation of the two pain responses in asphyxia is due to an action on the nerve trunk or

upon the nerve ending. The former is the case, the 1st response being abolished by asphyxia of the nerve trunk only.

Dissociation can also be accomplished by the use of cocaine.

Dissociation by cocaine. Cocaine reduces and ultimately abolishes the 2nd pain response, before it similarly affects the 1st pain response. A convenient method is to ionise the alkaloid into the skin of the front of the forearm just above the wrist, using a 0·5 or 1% solution of cocaine hydrochloride and a current of about 70 to 100 microamp. per sq. cm.; the effect is more certain (and may be necessary if the finger is used) if the circulation to the limb is stopped during the process, so that the cocaine entering the skin may be held in place. Diminished response to needle pricks develops obviously within 5 min. and it is easy to show that the 2nd pain response is affected first. It is lost before the 1st response, and often before the latter is much diminished. The cocainised skin gives a single and prompt response while uncocainised skin yields the usual pair of responses. This phenomenon is produced without difficulty. A similar phenomenon in response to the painful heat stimulus (the copper at 63°) can be demonstrated, though less easily. It is necessary to carry the cocainisation farther, and nearer to the point at which analgesia is complete, to reach the critical state in which the skin responds at once to brief contact with hot metal, but without the occurrence of the delayed sting. When the 2nd response to heat is lost, the 1st response is often much reduced, but sometimes is felt fully. The most certain way of eliminating the 2nd response only, is to cocainise until no response can be obtained by needle prick, and then to employ as stimulus brief contacts with the copper at 70°.

A similar differentiation can be elicited by infiltrating a superficial cutaneous nerve with a 0·1% solution of cocaine hydrochloride containing a small quantity of adrenaline. Using a cutaneous nerve of the forearm for this purpose, as little as 1 c.c. will suffice if it is introduced directly over the nerve trunk, and the weak solution gives a very slowly developing paralysis of sensation. Diminution and loss of the 2nd pain response to the needle prick is readily distinguished as analgesia develops, but the 1st response continues much longer. Here again paralysis must proceed farther before the corresponding phenomenon in response to the heat stimulus becomes clear, and this critical phase is brief, soon passing on to complete analgesia.

Discussion and further observations. Because the two pain responses can be dissociated by asphyxia or cocainisation that is confined to the nerve trunk, it becomes clear that dissociation concerns nerve paths and not nerve endings and thus indicates that the two pain responses are carried through distinct systems of nerves.

Now Gasser and his colleagues, in their experiments on animals, have shown that the fibres of peripheral nerve trunks conduct impulses centrally at very different rates. Studied in this way the fibres separate themselves

into three groups, A, B, and C. He has further shown that cocaine, in acting on the nerve trunk, abolishes first the function of the slower conducting (his C) fibres and, using our method of asphyxiation, that this first abolishes the function of the faster conducting (his A and B) fibres. Cocaine has long been known to abolish pain sense before touch(6), and it has been shown in this laboratory that asphyxia abolishes touch before pain ; from evidence of this kind it was natural at first to suppose that specific sensory functions might be associated with fibres of different conduction rate. But the idea has not been found capable of proof, and Gasser has expressed the belief that pain overlaps and may be conveyed in both the B and C groups of fibres. His evidence for this is indirect, his observations having been carried out upon animals, and the presence of pain being judged by reflex phenomena.

The present position is that our observations upon actual pain responses in man and those of Gasser on animals are in very clear and suggestive accord ; for he finds fibres of fast conduction rate, and we find fibres conducting the 1st pain response, to be the more susceptible to asphyxia ; while he finds fibres of slow conduction, and we find fibres conducting the 2nd pain response, to be the more susceptible to cocaine. The idea, which emerges, that the two pain responses are carried by fibres of different conduction rate, can be submitted to decisive experiment in man. For, if it is correct, then the interval between the 1st and 2nd responses should increase with the length of nerve traversed, and, because of its slow conduction, the 2nd response should be delayed very notably when the nerve path is long. This is all quite easy to demonstrate. If heat, the most suitable stimulus, is used and the point of stimulation is moved in steps from the finger up the arm to the shoulder, the interval between the two responses gradually diminishes until near the shoulder the responses fuse. Similarly in the case of the leg ; when the foot is stimulated the interval between the responses is remarkable for its length,* and this interval decreases as the stimulus is moved upwards, until near the hip joint the two responses are indistinguishable.

The lag of the 2nd response from the foot is so great that if two heat stimuli are given in quick succession to the same skin, a double echo is felt after the copper has been removed from the skin. This phenomenon was also noted by Rosenbach. It is important because it would seem in itself to exclude the idea of any " between process " (*see* Thunberg) as accounting for appreciable delay between stimulus and response.

We have used a simple contrivance as did Thunberg to measure the interval between the stimulus and the 2nd response. Two thin copper wires are stretched in parallel and 5 mm. apart across an open frame as handle. The bare wires are laid on the skin to be tested and the hot copper completes a circuit the instant it touches them. The make and break of contact with the skin is thus signalled ; the pain response is signalled by

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* Gad and Goldscheider (4) noticed the greater delay of pain response when stimulation of the foot was compared with that of the hand.

depressing a key which completes the same circuit. An example of the records is shown in Fig. 2, and representative values are given in Table II.

From these values the rates of conduction of the 2nd response can be calculated and, owing to the magnitude of the intervals involved, without
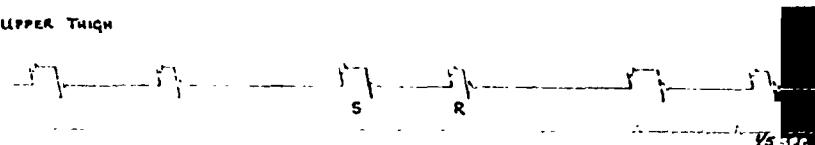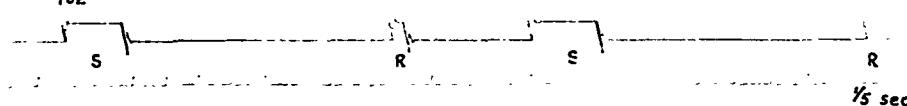

Fig. 2. (× ¾.) Measurements of heat stimulus to 2nd pain response, the stimulus being applied to the upper thigh and to the toe. *S* signals the stimulus and *R* the pain response. Time in 1/5 sec..

TABLE II.

Stimulus to 2nd response in sec..

	1st series.			2nd series.		
	Thigh.	Knee.	Toe.	Toe.	Knee.	Thigh.
Subject T.L.	0·8	1·1	1·9	1·8	1·2	0·8
(reaction time 0·2).	0·8	1·1	2·0	2·0	1·2	0·8
	0·7	1·1	2·1	.2·0	1·2	0·8
	0·7	1·1	2·0	1·9	1·3	0·8
Av. in sec.	0·8	1·1	2·0	1·9	1·2	0·8

Thigh to knee 40 cm., knee to toe 40 cm., the points being previously so arranged along the outer side of the leg.
Calculated rate of conduction, toe to knee, 0·5 metres per sec..
   „   „   „   toe to thigh, 0·7 metres per sec..

	1st series.			2nd series.		
	Toe.	Knee.	Thigh.	Toe.	Knee.	Thigh.
Subject E.E.P.	2·0	1·5	1·0	1·7	1·2	0·9
(reaction time 0·15)	1·9	1·4	0·9	1·8	1·4	0·9
	2·0	1·4	1·0	1·8	1·5	0·9
	1·8	1·3	0·9	1·8	1·4	0·9
Av. in sec.	1·9	1·4	1·0	1·8	1·4	0·9

Thigh to knee 50 cm., knee to toe 50 cm., the points being previously so arranged along the outer side of the leg.
Rate of conduction, toe to knee 1·1 metres per sec..
   „   „   toe to thigh 1·1 metres per sec..
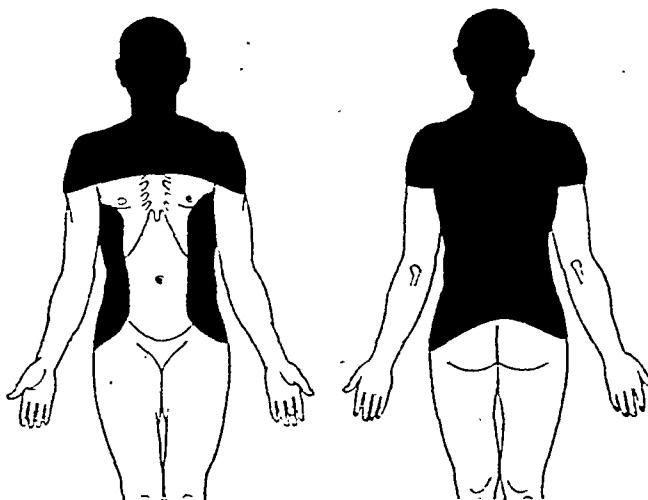
Fig. 3.
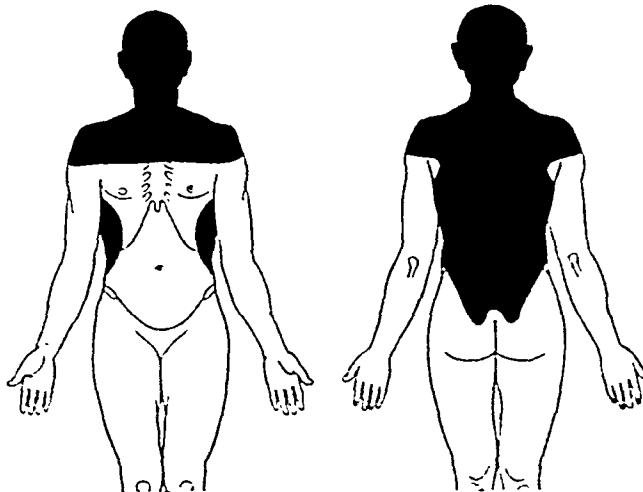
Fig. 4.

Figs. 3 and 4. Independent observations by the two authors, heights 5 ft. 8 inches and 6 ft.
respectively. The blackened areas are those over which heat gives a single pain response ;
on the unblackened areas the skin gives a double response, the interval between the two
increasing as the stimulus is moved peripherally on arm or leg.

gross error.* They are about $\frac{1}{2}$ to 1 metre per second. These are of course approximate values; but they are of interest in being of the same order (1 to 2 metres per sec.) as those found by Gasser for his C fibres.

It has been demonstrated that difference in conduction rate is responsible for the double pain response; it remains to be shown that the lag of the 2nd impulse is not appreciably influenced, as Gad and Goldscheider suppose, by its passage through spinal cord. If the heat stimulus is used and the whole surface of the body is tested, the double response is found to be unobtainable from the blackened areas of Figs. 3 and 4. It is obtained from all other parts, but shortens until it is unrecognisable wherever the areas indicated in black are closely approached.† It will be manifest from the diagrams that the cord can contribute little‡ to the lag of one pain response behind the other, for the boundaries at which the double response fuses into one everywhere stand at approximately the same distance, measured along nerve trunks to their points of entrance into the cord; and this is so, whether the measurement is to the cervical or to the sacral region of the cord.

The evidence, derived from man alone, seems definitely to show that pain is transmitted from the skin by two sets of nerve fibres, of fast and slow conduction. Taken in conjunction with the earlier work of Gasser the truth of this conclusion is all the more certain. It is of interest to note that relying upon pain itself as our index, the incidence of the pain nerves of different conduction rate is not a diffuse scatter or transition from fast to slow, the fibres must be almost if not quite sharply separated into a fast and a very slow conducting group.

These observations may prove relevant to the interpretation of the delay in appreciation of pain which occurs in cases of spinal cord disease, such as tabes dorsalis.

It is also to be remarked that having distinguished between pain impulses carried by slow and fast fibres, it becomes necessary to reinvestigate pain sense from this standpoint. It is possible, or in some instances even probable, for example, that there are two corresponding series of nerve endings, that the thresholds of these are different, and that when the nerve fibres are cut, they regenerate at different rates.

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* One possible source of error, though it cannot be of great magnitude, arises out of the response starting within different spinal nerve territories, thus having to travel to different cord levels. Another is the thickness of the skin stimulated.

† Tested with heat, the face, like the rest of the areas represented black, yields a single response. A needle prick of the face is often followed by a second sensation, which comes gradually and lasts for very many seconds. It is a little sense of continuous sting, the nature of which we have not investigated, but which is to be distinguished from the fleeting pain of a second response as this is felt in finger or toe.

‡ We do not pretend to have proved that it contributes nothing.

Additional observations.

1. *A correlation.* It may be of value here to remark upon a correlation, which is not directly relevant to the conclusions of this paper. Asphyxia affects the functions of the sensory nerves in a given order, cocaine affects these same functions in the reverse order. It is becoming a matter of consent that these differences are due to differences in the constitution of the nerve fibres concerned. In an earlier paper it was suggested that the paralysis occasioned by asphyxia is centripetal for similar reasons, fibres supplying distant skin being regarded as differently constituted to those supplying less distant skin. We are inclined here to emphasise this view for closer consideration, because the interesting phenomenon of centripetal paralysis has received no other explanation than that which was put forward, and because Endres (3) has shown that cocaine has the reverse effect, producing centrifugal paralysis. This effect of cocaine we have confirmed in the present observations. If, in comparing the paralytic action of asphyxia and cocaine, we attribute the reversal of one series of events to differences in fibre constitution, it is difficult to avoid attributing reversal of a second series of events to a similar cause. The importance of this consideration is that it affects the interpretation of physiological experiments on the different rates of nerve fibre conduction.

2. *Pain localisation.* In a previous paper from this laboratory (7) observations were described showing that painful stimuli are more or less accurately localised by the subject. To avoid the error arising out of the presence of touch nerves, these were thrown out of action by asphyxia, the observations being made on anæsthetic skin. In the light of the present work it will be evident that the localisation of pain thus investigated applies only to that conveyed by the group of slowly conducting nerve fibres.

We have now examined the power to localise pain conveyed by fast conducting fibres. For this we have used single induction shocks thrown at each separate test into one of two pairs of electrodes, both resting on the skin. These induction shocks awaken the 1st pain response only and the observer attempts to identify the electrodes through which the stimulus is received. He has no difficulty in doing so when the two pairs of electrodes lie across the skin of the forearm separated by distances exceeding 1 cm. and when they lie up and down the arm separated by distances exceeding 1·5 cm.. Thus localisation is of much the same order of accuracy whether the 1st or the 2nd pain response is considered.

Conclusions.

It is demonstrated that the human skin is supplied by pain nerves having very different conduction properties. The incidence of fibres of different conduction rate is not a diffuse scatter or simple transition from fast to slow, the fibres are more or less sharply divided into a fast and a very

slow conducting group. It is for this reason that a single stimulus such as a needle prick or heat is capable of giving two distinct pain responses, as described by Thunberg and others. Both these pain responses are localised by the subject.

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THE ACTION OF CHOLINE ESTERS IN MYASTHENIA GRAVIS.*

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Introduction.

THE cause of the weakness of the voluntary muscles in myasthenia gravis is unknown, and no satisfactory explanation for the fatigue phenomenon characteristic of this disease has been offered. The administration of a number of substances, such as ephedrine (10, 11), glycine (2) and potassium salts (19), has been found to produce improvement in the muscle power, and in 1934 Walker (34) demonstrated that physostigmine when given by hypodermic injection has a powerful action in remedying the weakness and fatigue. When given in sufficiently large doses to be fully effective, however, this drug has undesirable toxic effects and Walker (35) showed that the injection of prostigmin, a synthetic analogue of physostigmine, can produce dramatic recovery in doses that do not produce such severe toxic actions. These toxic actions, especially slowing of the heart and increased intestinal peristalsis, can be prevented by atropine (9). When given by the mouth in much larger doses than those effective by intramuscular or hypodermic injection prostigmin is effective (20), but by either method of administration the action is temporary and persists for a few hours only. The therapeutic effect of prostigmin has been confirmed by numerous observers (29, 18, 21, 27, 33, 26, 5, 37). This remedial action has renewed interest in the problem of the pathology of myasthenia gravis, for many of the effects of physostigmine and of prostigmin can be related to their action in inhibiting the choline esterase of the body, which rapidly hydrolyses acetyl choline (6, 22, 23, 24, 12), and acetyl choline is now known to be necessary for the transmission of nerve impulses in sympathetic ganglia (13), from nerve endings to the effector organs of the parasympathetic system (7), and also from voluntary nerve fibres to skeletal muscles (8) (3). These facts suggest that in myasthenia gravis the normal balance between the production of acetyl choline at the neuro-muscular junction and its destruction by the esterase is disturbed.

The esterase content of the serum in patients with myasthenia gravis has been studied and is found to be within the normal range (32, 14, 25). It is possible, as pointed out by McGeorge, that the esterase activity at the neuro-muscular junction may be disturbed in the presence of a normal serum esterase activity, but so far no method of testing this hypothesis has been devised. A second possibility is that there is some abnormality in the formation of acetyl choline. Walker (36), and Cooke and Passmore (5) tried the effect of the subcutaneous injection of 50 and 100 mg. of acetyl choline chloride in patients with myasthenia gravis, but observed no effect. In view of the rapid destruction of acetyl choline in the body, this is not surprising and we considered that further evidence might be obtained by observing the effects of the more stable choline esters which have actions similar to those of acetyl choline. Two types of actions of acetyl choline can be distinguished, muscarine-like or parasympathomimetic actions, which can be prevented or abolished by atropine, and nicotine-like actions that are not affected by atropine (6). The action of acetyl choline in the transmission of voluntary nerve impulses to skeletal muscles belongs to the nicotine-like group.

Observations.

We were fortunate in being able to obtain two patients from Dr. Walker whom she had observed for two years and on whom she had made her observations of the effects of treatment by prostigmin. (Case reports are appended). Both patients were so severely affected by the disease, the muscles of the limbs and trunk, as well as of the face and eyes, being involved, that the effects of treatment were easily gauged.

The procedure adopted was to stop routine treatment with prostigmin following the evening or afternoon dose and by next afternoon the patients were so weak that the eyelids could scarcely be stirred, the external ocular movements were almost completely absent, the head could, as a rule, not be turned or raised from the pillow, the ankles could not be dorsiflexed or the heels raised from the bed, and sitting up and walking were impossible. One of the patients (W.) could still raise the arms from the bed, but her hand grips were very weak, while in the other (C.) the upper limbs were as weak as the rest of the body. By observing the ability to perform these movements and by using a small dynamometer to gauge the power of the hand grips, the return of muscle power could readily be observed and its extent recorded. Readings of pulse rate and blood pressure were taken every two or three minutes, and the choline ester was given by subcutaneous injection after the preliminary observations had been continued for from 15 to 30 minutes.

Subcutaneous injection.—Carbaminoyl choline chloride (Doryl) was first tried and in doses of 1·0 mg. produced a striking result (*see* Figs 1 *a* and *b*, and 2 *a* and *b*). · Compared with the effect of prostigmin the return of muscle power commenced later (*see* Tables I and II), developed more slowly, was

never so complete, but lasted much longer. Whereas on routine treatment with prostigmin (three injections of 2·5 mg. in one day) the patients were constantly too weak next morning to attend to themselves, following carbaminoyl choline they could move freely in bed and sometimes even sit up. Patient C. always developed abdominal pains of a colicky nature following carbaminoyl choline, as she did after prostigmin, and she was given a preliminary injection of 1·0 mg. of atropine sulphate which controlled them ; patient W. with doses of 1·0 mg. or more developed excessive salivation, lachrymation, sweating and fall of blood pressure, which could not be entirely controlled by atropine, but with 0·5 mg. these were not conspicuous. In all, patient C. was given nine subcutaneous or intra-muscular injections of carbaminoyl choline in doses of 0·5 to 2·0 mg., and patient W. eleven injections in doses of 0·25 to 2·0 mg., and the effects on muscle power were essentially the same on each occasion.

Acetyl choline chloride in doses of 500 mg. and 600 mg. produced a definite recovery of muscle power, not nearly so satisfactory as that following carbaminoyl choline, but similarly delayed and still present next morning. No parasympathomimetic effects were observed apart from a slight tightness in the chest in patient W., and abdominal colic in patient C. a few minutes after the injection.

Acetyl β-methyl choline chloride (Mecholin) in doses of 25 mg. and 50 mg. produced good recovery of muscle power similarly delayed and noticeable next morning. Compared with acetyl choline and carbaminoyl choline, it is regarded as feeble in nicotine-like effects though powerful in " muscarine " effects (31), and little if any result on voluntary muscle power was expected (1). In both patients atropine was required to control the cardio-vascular effects.

In view of the very rapid destruction of acetyl choline in the body and the prolonged effects on muscle power that resulted from the injection of these esters, it seemed possible that the choline contained in them was being utilised in the synthesis of acetyl choline, or its precursor, at the neuro-muscular junctions. Injections of choline chloride in doses of 200 mg. were, however, without effect, even after the oral administration of sodium acetate in doses of 6·0 grammes daily for three days, as was also the oral administration of 3·0 grammes of choline chloride and 6·0 grammes of sodium acetate daily for a week. To test this possibility further, the following observations were made. The three esters contain practically the same amount of choline so the effect of 1·0 mg. of each was observed. The results were widely different ; 1·0 mg. of carbaminoyl choline produced a conspicuous effect, 1·0 mg. of acetyl β-methyl choline a weaker effect, while 1·0 mg. of acetyl choline produced no recognisable effect. The actions of the esters could not, therefore, be due merely to their choline content. Tables I and II show the comparative effects of prostigmin, the three esters and choline.

TABLE I. (Patient C.).

Treatment.	Effect evident.	Sitting up.	Walking.	Effect ended.
Prostigmin 2·5 mg.	3 mins.	5 mins.	8 mins.	6 hrs.
Carbaminoyl choline (doryl) 1 mg.	8 mins.	2 hrs.	—	> 20 hrs.
Acetyl choline 200 mg. 600 mg.	9 mins.	2 hrs.	—	> 18 hrs.
Acetyl β-methyl choline (mecholin) 25 mg.	5 mins.	70 mins.	80 mins.	> 18 hrs.
Choline 200 mg.	—	—	—	—

TABLE II. (Patient W.).

Treatment.	Effect evident.	Sitting up.	Walking.	Effect ended.
Prostigmin 2·5 mg.	5 mins.	8 mins.	9 mins.	6 hrs.
Carbaminoyl choline (doryl) 0·5 mg.	3 mins.	55 mins.	3½ hrs.	> 17 hrs.
Acetyl choline 500 mg.	90 mins.	—	—	> 17 hrs.
Acetyl β-methyl choline (mecholin) 50 mg.	18 mins.	12 hrs.	12 hrs.	> 15 hrs.
Choline 200 mg.		—	—	—

Intra-arterial injection. In spite of the rapidity with which acetyl choline is destroyed by the esterase of the body, it was possible that small amounts of the three esters were entering the circulation and, escaping hydrolysis, were reaching the neuro-muscular junctions; so an attempt was made to supply them more directly to the muscles by intra-arterial injection. 100 mg. of acetyl choline chloride injected rapidly into the right femoral artery of patient W. produced within 2 seconds a bright red flush in the skin throughout the distribution of the artery, but no change in muscle power occurred until 2 hours later, when there was some improvement in the musculature of the whole body, and the legs could be moved freely and raised from the bed, but it was no greater in the leg injected than elsewhere (Table III). 5 mg. of acetyl β-methyl choline chloride were injected into the right femoral artery, a sphygmomanometer cuff being distended at a pressure of 80 mm. of Hg around the thigh to delay the diffusion of the drug throughout

TABLE III. (Patient W.).

Effect of acetyl choline chloride by intra-arterial injection.

Time.	
0	Acetyl choline chloride 100 mg. in 0·5 c.c. into right femoral artery. Immediate pain in thigh and leg to sole of foot.
2 secs.	Bright flush throughout cutaneous distribution of femoral artery. A few beads of sweat on toes.
2 mins.	Flush fading.
12 mins.	Flush disappeared. No change in muscle power.
2 hrs.	Muscle power improved, but no difference between the two legs.
19 hrs.	Next morning a little stronger than usual.

the body, but no immediate effect was produced on muscular power. The pressure in the cuff was released at the end of six minutes, and thirteen minutes following the injection the external ocular muscles and the muscles of the limbs became stronger, and she was able to sit up one hour and walk two and a half hours after the injection. No difference was noted between the strength of the muscles in the two legs. Using a similar technique 0·25 mg. of carbaminoyl choline was injected into the left femoral artery and three minutes later the muscles of the injected leg showed a definite, but slight, return of power, so that she could move the toes and dorsiflex the ankle. No further improvement had occurred when the cuff was released seven minutes after the injection. Twenty-four minutes after the injection a general improvement in the muscles of the body commenced, but the injected leg was still stronger than the other. Forty-five minutes after the injection she was able to sit up and the two legs were equally strong, and at the end of one and a half hours she could walk (Table IV).

TABLE IV. (Patient W.).

Effect of carbaminoyl choline chloride by intra-arterial injection.

Atropine sulphate 1·2 mg. by subcutaneous injection ½ hour previously.

Time.	
0	Carbaminoyl choline chloride 0·25 mg. in 0·5 c.c. into left femoral artery with venous compression in middle of thigh.
3 mins.	Able to move toes and ankle on left side.
7 mins.	Venous compression released.
24 mins.	Able to move toes and ankle on right side. Eyelids raised. Both arms recovering.
27 mins.	Legs equally strong.
41 mins.	Sitting up.
1½ hrs.	Can walk a few steps.
18 hrs.	Next morning stronger than usual.

These results suggested the possibility that the defect in myasthenia gravis and the beneficial effects of prostigmin and of the choline esters might be situated more centrally than the neuro-muscular junctions. To test this, 1·0 mg. of prostigmin was injected into the left femoral artery of the same patient, compressing the thigh with the sphygmomanometer cuff at a pressure

of 80 mm. of Hg as before.   Two minutes later the toes and foot twitched involuntarily and in three minutes strong voluntary movements were possible. At the end of five minutes the heel could be raised from the bed, but no muscle power had returned in the other leg or the body generally.   At the end of seven minutes the pressure in the cuff was released and two minutes later the muscle power was returning throughout the body.   Five minutes after the release of the pressure the patient was sitting up, but the injected leg was still the stronger, and one minute later she was walking and the two legs appeared to have recovered equally (Table V).

TABLE V.   (Patient W.).

Effect of prostigmin by intra-arterial injection.

TIME.	
0	Prostigmin 1·0 mg. in 0·5 c.c. into left femoral artery with venous compression in middle of thigh.
2 mins.	Toes and ankles twitching involuntarily.
3 mins.	Strong dorsi and plantar flexion of ankle.
5 mins.	Heel raised from bed, no improvement on right side.
7 mins.	Venous compression released.
9 mins.	Strength returning throughout body.
12 mins.	Right leg raised from bed, not so strong as left. Sitting up.
13 mins.	Walking. Right leg as strong as left.

Discussion.

The immediate and local effect of prostigmin by intra-arterial injection and the rapid general recovery of muscle power when the drug was allowed to enter the general circulation, point to a peripheral site of the lesion in myasthenia gravis—probably the neuro-muscular junctions—and so support the conclusion of Pritchard (29) who studied the form of the myogram in this disease.   The delayed effect of the choline esters when given by the same route makes it difficult to believe that they acted by being conveyed directly to the neuro-muscular junctions, except in the case of carbaminoyl choline, when slight immediate recovery occurred in the injected leg, to be followed later by the delayed recovery in the muscles of the body generally, similar to that following acetyl β-methyl choline and acetyl choline.   Carbaminoyl choline is much more stable than acetyl choline (17) and it is probable that the immediate effect of the former was the result of some of the drug injected reaching the neuro-muscular junction before it was hydrolysed.   Acetyl β-methyl choline is also more stable than acetyl choline, but has little nicotine-like action and when given by quick intra-arterial injection has no muscle stimulating action in the cat similar to that of acetyl choline, and so would not be expected to act, even if it reached the neuro-muscular junction (1).   In the case of acetyl choline, the vasodilatation that followed immediately was evidence that the drug had reached the surface capillaries before it was hydrolysed, but the difficulty of applying it sufficiently rapidly and

directly to the neuro-muscular junction to affect voluntary muscle before it is hydrolysed, has been pointed out by Brown, Dale and Feldberg (3). The delayed effects of each of the esters following intra-arterial injection were essentially the same as those that followed their administration by subcutaneous injection, and were similar for each of the three esters. A gradual absorption of small amounts of the esters from the site of subcutaneous injection and their conveyance as such to the neuro-muscular junction is not supported by these results. Other possibilities that could account for their effects are (1) that they have a more central action on the nervous system, or (2) that they are utilised after absorption and hydrolysis for the synthesis of acetyl choline, or (3) that they are utilised after absorption for the elaboration of a precursor which breaks down to form acetyl choline on the arrival of the nerve impulse at the neuro-muscular junction. A fourth possibility, that the administration of the choline esters produces a lowering of the esterase activity similar to the action of prostigmin, can be eliminated, for Hall, Ettinger and Banting (15) found no significant alteration in the esterase activity in the serum of dogs on prolonged administration of acetyl choline, and we have found no change following the injection of carbaminoyl choline in two healthy adults, who experienced colicky abominal pains at the time the esterase activity of the serum was estimated, showing that the effects of the drug were then present. We have also found that the esterase activity of serum is not depressed by the addition in vitro of carbaminoyl choline and acetyl β-methyl choline in concentrations much greater than could occur in the body following the injection of effective doses.

In considering these hypotheses account must be taken of the quantitative differences in the effects of the three esters, these effects being in the order of the stability of the esters. Their action must, therefore, be due to their presence in the body as esters and not merely as sources of choline, the common product of their hydrolysis ; and the failure to observe any effect from the injections or oral administration of choline is evidence in support of this. That the esters are utilised, after hydrolysis, in the synthesis of acetyl choline is, therefore, improbable.

That acetyl choline has an action on the central nervous system has been shown (16, 30), but these effects are inhibited by atropine, and in our observations the effects were independent of whether atropine was given or not. Further, no evidence of a central lesion in myasthenia gravis has been produced. Acetyl choline has a stimulant effect on sympathetic ganglia, which is not inhibited by atropine ; the possibility of the choline esters acting through this mechanism has been considered, especially in view of the beneficial effects of ephedrine in this disease. Both patients derive benefit from the oral administration of ephedrine, and believe that it potentiates and prolongs the effects of prostigmin. In patient W. intramuscular injections of adrenaline (0·5 mg.), ephedrine (64 mg.) and of benzedrine (25 mg.) had no effect on the myasthenia, but in patient C. both ephedrine and benzedrine produced delayed and prolonged recovery similar to that

immobile with the lips slightly parted, and they could be retracted a little further. The left eyelid could not be raised, the right a quarter way. The pupils were moderately dilated, reacted to light, but not to attempted convergence ; the eyeballs were immobile and tears collected in the conjunctival sacs. No contraction of the masseters was perceptible. The tip of the tongue could be protruded between the teeth, the palate was raised on phonation, swallowing was possible but tired rapidly, the voice was feeble. Shrugging of the shoulders was just possible, and the head was scarcely able to be turned from side to side. The respiratory muscles were feeble and mainly diaphragmatic. All movements of the upper limbs were possible but very weak, the biceps jerks were present but weak, the other reflexes absent. The abdominal reflexes were present but weak, and the muscles of the abdominal wall could be felt to contract on attempting to sit up, but this could not be repeated. No contraction of the muscles of the lower limbs was observed, but the tendon reflexes were present, though feeble. Superficial sensations were normal. Micturition and defæcation were possible but difficult. X-ray examination failed to show enlargement of the thymus.

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Fig. 2.

Fig. 2. Patient C. Two hours after subcutaneous injection of 1·0 mg. carbaminoyl-choline chloride. (doryl).
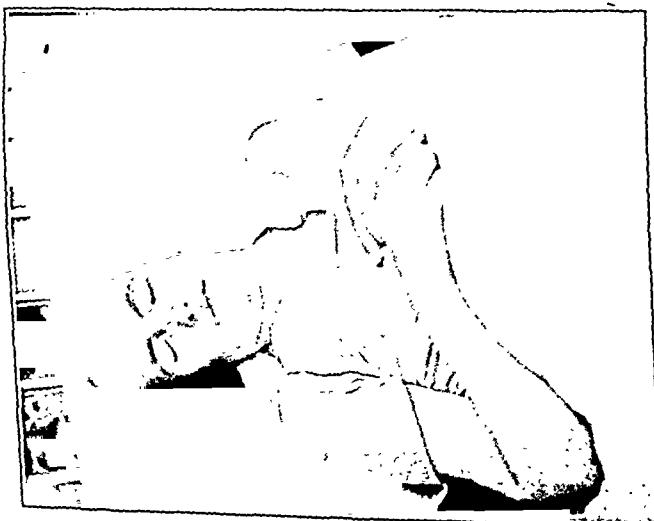

Fig. 1.

Fig. 1. Patient C. All treatment suspended for sixteen hours.

Fig. 4. Patient W. Two hours after subcutaneous injection of 0·5 mg. carlaminoyl choline chloride (doryl).

Fig. 3. Patient W. All treatment suspended for twenty-four hours.

INSULIN RESISTANCE AND THE ARTERIO-VENOUS BLOOD-SUGAR DIFFERENCE.

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WE are passing through a phase in the attack on the problem of carbohydrate metabolism which ultimately may well prove to be as fruitful as the phase which resulted in the discovery of insulin. The pituitary gland, long known to play a major part in the co-ordination of growth and metabolism, and to be concerned also with sugar utilisation in syndromes such as those of Cushing and Fröhlich, is the focus of interest at the present time. Investigations on the relation of the pituitary to carbohydrate metabolism, which in their present aspect originated in the work of Houssay and his colleagues, have been described in detail by Collip (5) and others (38 and 40), and it will suffice for us to indicate briefly those points of interest which more directly bear upon our subject. It has been shown (*a*) that removal of the anterior lobe of the pituitary gland from the depancreatised animal ameliorates the diabetic condition (23); (*b*) that the hypophysectomised but otherwise normal animal is hypersensitive to insulin (8, 16 and 25); and (*c*) that injection of extracts of the anterior pituitary produce resistance to the hypoglycæmic action of insulin, and even hyperglycæmia and glycosuria (1, 2, 6, 10, 24, 31, 37 and 39). These researches afford convincing evidence not only that the diabetic symptoms of the depancreatised animal are, in part at least, due to the so-called diabetogenic influence of the pituitary, but also that an excess of this pituitary principle in the blood of an animal can produce hyperglycæmia, glycosuria and resistance to the action of insulin. The application of this information, derived from animal experiments, to the problem of diabetes in the human subject is at once desirable and full of promise.

The existence amongst diabetic subjects of a group in which it seemed possible that pituitary dysfunction rather than insular insufficiency might be the ætiological factor, led Prof. De Wesselow and myself (9) to examine the blood-plasma of such patients for the presence of a pituitary-like

substance. Using as a test the hypoglycæmic response of the rabbit to insulin given intravenously, we were able to show that previous injection of the plasma of certain elderly diabetics diminished the duration of the hypoglycæmia, whereas the blood of young diabetics was uniformly without this effect. It appeared to us that these findings are against the view that disturbances of carbohydrate metabolism—apart from obvious pituitary conditions—are invariably due to a lack of insulin, and that their close similarity to those obtained by other workers with extracts of the anterior pituitary suggests that the blood of certain elderly diabetics contains a pituitary-like diabetogenic factor.

The phenomenon of insulin resistance in diabetes is of interest in view of the effect of pituitary extracts on the insulin response. In the early days of insulin therapy temporary resistance to the hormone was frequently noted to accompany infectious conditions ; also cases of unexplained resistance were recorded (see Lawrence (30)). Resistance to the action of insulin with no obvious cause is now well recognised, and during the last few years attempts have been made to classify diabetics according to the ease with which they respond to insulin ; Falta (11), Himsworth (21) and MacBryde (32) have recorded the existence of insulin-sensitive and resistant patients. That the blood of certain elderly diabetics often produces in rabbits a kind of resistance to insulin raises the question of the type of response which such subjects would show to injected insulin. We therefore decided to investigate the reaction of the two classes of diabetic, the young and the elderly, to insulin. Further, we thought it would be of interest to study the behaviour of the arterio-venous blood-sugar difference (A.V. difference) after glucose and insulin in the two groups, since Himsworth (21) has stated that resistance to insulin is accompanied by an impaired peripheral power of sugar utilisation.

Experimental.

Because the effects of insulin on the blood-sugar level in different subjects can be compared only when the initial blood-sugar figures do not greatly differ from each other, the insulin depression curve cannot in general be used to assess insulin sensitivity. We have, therefore, used the convenient method described by Himsworth (21), which records the efficiency with which insulin is able to control the hyperglycæmia following ingestion of glucose. The patient received no food or insulin after supper on the evening preceding the morning on which the test was carried out. After removing samples of blood for determination of the fasting sugar content, 5 units of insulin were injected intravenously and immediately afterwards the subject was given 50 g. of glucose in 100 c.c. of water. Blood samples for sugar estimation were taken 30, 60 and 90 min. after injection of the insulin. The patients were confined to bed on the morning of the test.

The sugar of arterial and venous blood was estimated by MacLean's method on 0·1 c.c. of blood. Arterial blood was obtained by *deep* puncture

(3 mm.) of the *pulp* of the thumb or finger with a spring lancet; the first drop of blood was wiped away and a 0·1 c.c. pipette filled with the blood as it issued from the wound. If at the beginning of the experiment the hand was cold, it was warmed in hot water for a few minutes, and in order to maintain as far as possible uniform conditions the patient was instructed to keep the arm from which the blood samples were to be taken under the bedclothes and close to the body during the intervals between bleeding. Usually the blood was easily obtained without mechanical aid, but occasionally it was necessary to employ gentle massage; a tourniquet was never used. Immediately the arterial sample was obtained we proceeded to withdraw some venous blood. After sterilising the skin with ether a tourniquet was applied to the arm and without loss of time an antecubital vein was pierced with a fine needle attached to a small syringe and about 0·2 c.c. of blood withdrawn. The blood was immediately transferred to a small vessel and 0·1 c.c. taken up in a pipette before sedimentation or clotting could occur; no anti-coagulant was used. The use of a tourniquet greatly facilitates the venepuncture, and by independent tests we were satisfied that momentary stasis does not influence the blood-sugar. All samples were withdrawn from the same vein. As the entire operation may be performed in a little more than a minute the interval between drawing the arterial and venous blood samples can be ignored. The following patients were studied :—

1. *Controls.* Six subjects of various ages; 3 males and 3 females, suffering from various chronic diseases but with normal carbohydrate metabolism, and on an ordinary mixed diet.

2. *Young diabetic subjects.* Six females, of whom 5 were under thirty, while in the remaining case the disease was detected at the age of 49. They were on carbohydrate rations of from 70-100 g. per day. with the exception of 2, who were receiving 190 and 240 g.. All were having insulin.

3. *Elderly diabetic subjects.* Six females, between the ages of 51 and 61, on carbohydrate rations of 70-100 g. per day. One only was taking insulin. The average weights were as follows : controls, 9 st.; young diabetics, 8 st.; elderly diabetics, 10 st.. The cases were taken without selection, apart from age, from those admitted to our wards for stabilisation.

Himsworth (21) deemed it desirable for comparative purposes that the patients should all be receiving approximately the same amount of carbohydrate. In all of our patients but two, who had been stabilised on rather high carbohydrate rations, this condition was observed.

Results.

The effect of insulin on the arterial blood-sugar. In the normal subject the effect of insulin on the hyperglycæmia resulting from the ingestion of glucose—the so-called alimentary hyperglycæmia—was apparent in both arterial and venous values, but it will be advantageous to describe, in the first place, the changes which occurred in the arterial blood-sugar. since

these changes show the degree to which insulin was able to suppress the alimentary hyperglycæmia, which, according to Himsworth (21), is a measure of the sensitivity of the individual to insulin.

The curves obtained from the normal subjects after glucose, both with and without insulin, were averaged and are shown in Fig 1A. Insulin caused a definite suppression of the hyperglycæmia, particularly at the 30 min. period after its injection. A second depression of the blood-sugar, which occurred 90 min. after insulin, was most probably due to endogenous insulin secreted in response to the rise in blood-sugar which followed the initial depression; this fall in the blood-sugar at the 90 min. interval was a constant feature of the curves.
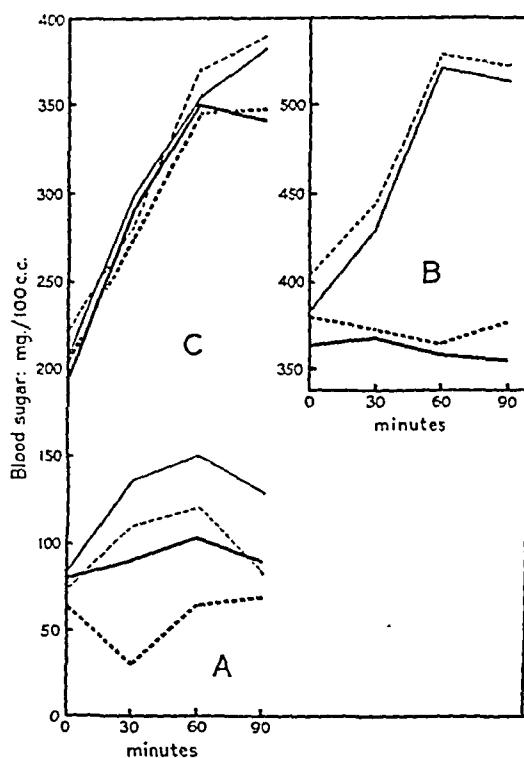
Fig. 1. Average arterio-venous blood sugar curves from 6 cases in each group : A, normals; B, insulin-sensitive diabetics ; C, insulin-resistant diabetics.

Continuous line = arterial blood sugar.   Dotted line = venous blood sugar.
Faint lines = glucose alone.   Heavy lines = glucose and insulin.

Inspection of the curves obtained on the young diabetics, examples of which are given in Figs 2, 3 and 4, shows that in every case the suppression of the blood-sugar by insulin was marked, so that the average curve of the series (Fig. 1B) shows complete abolition of the alimentary hyperglycæmia, which after glucose alone averaged 130 mg./100 c.c.. It will readily be appreciated that the hypoglycæmic action of insulin in these young patients was at least equal to that observed in the normal patients ; in other words,

they were normally sensitive to insulin. Although it might appear that they were more sensitive than the normal, it must be remembered that the high initial blood-sugar value is a factor which contributes to the effect of insulin.

In the case of the elderly diabetics inspection of the results, typically represented in Figs 5 and 6, reveals at once that the response of the arterial blood-sugar to insulin, so pronounced in the other groups, was almost negligible. This finding affords the clearest evidence that these patients were resistant to insulin. In a few cases we tried the effect of giving larger doses of insulin : we found that in this way it was possible only to diminish
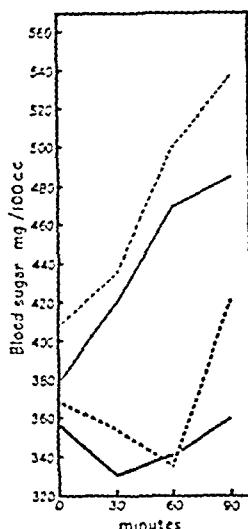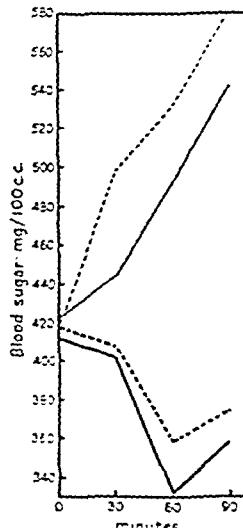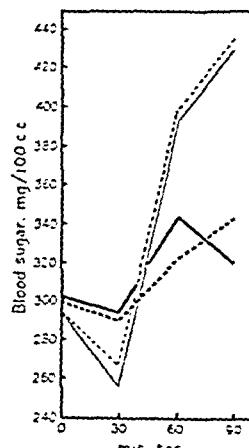
Fig. 2.   Fig. 3.   Fig. 4.

Young diabetics : curves showing sensitivity to insulin.

the blood-sugar rise by a relatively small extent. It will be observed that the sudden changes in the level of the blood-sugar, which give to the curves of the other groups a certain irregularity, are absent from the curves of the elderly patients, a finding which suggests a persistent and unyielding resistance to the action of insulin.

The effect of insulin on the A.V. difference. The average arterial and venous blood-sugar curves of normal subjects after glucose alone, and after glucose and insulin, are shown in Fig. 1A. The A.V. differences which we obtained in these subjects under fasting conditions showed wide variations, but in all cases the arterial blood-sugar was at least equal to, and was often higher than, the venous value. The maximum difference was 40 mg./100 c.c., and the average value 11 mg./100 c.c. of blood. These figures correspond with those of Friedenson and his co-workers (15) for normal students ;

other workers have recorded small positive differences with the exception of Grott (18), who obtained a few negative differences in normal fasting subjects. We wish strongly to emphasise the almost complete absence of negative A.V. differences from our results on normals since, as we shall show in the next section, such negative differences occurred with some frequency amongst the diabetic findings. The increase in the A.V. difference after ingestion of glucose is well-known, and is due to the peripheral action of insulin secreted in response to the rising blood-sugar. The effect of injected insulin on the A.V. difference in the normal was pronounced, and showed its maximum usually at the 30 min. period. It is possible, therefore,
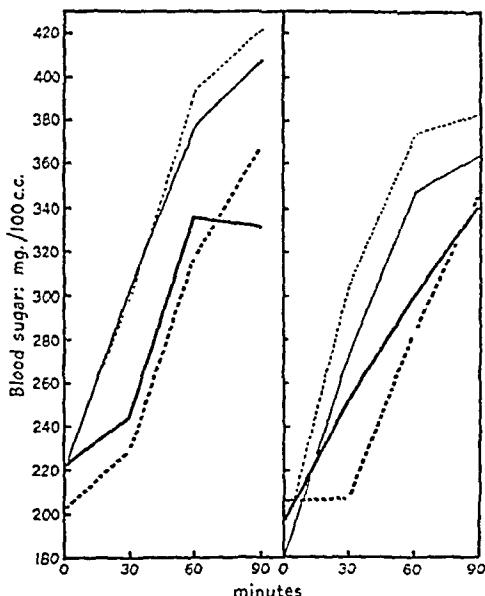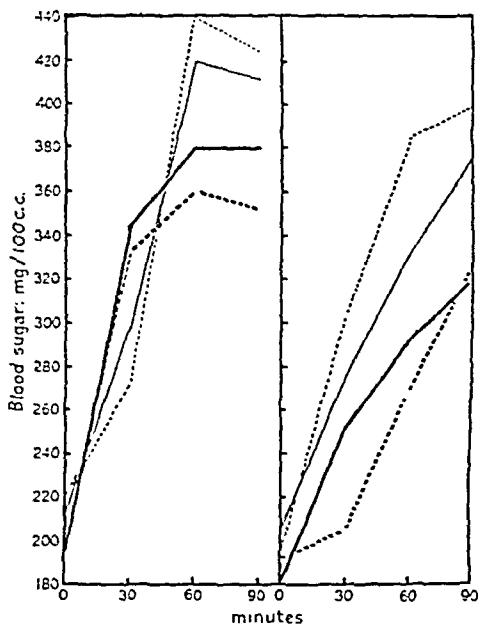
Fig. 5.   Fig. 6.

Elderly diabetics : curves showing resistance to insulin.

to demonstrate a two-fold effect of insulin given with glucose to the normal subject : (1) a suppression of the arterial hyperglycæmia, and (2) an increase in the A.V. difference ; these effects are manifested during the hour following injection of insulin.

In our studies the behaviour of the A.V. difference in the young diabetic (Figs 2, 3 and 4) has shown a striking departure from the normal. There was a tendency for the fasting A.V. difference to be negative, often to a considerable degree. Following glucose this negative difference was, in most cases, maintained ; there is, however, one outstanding exception in which an initial negative difference was converted to a positive value which persisted throughout the experiment. It is clear, therefore, that so far from removing sugar from the blood in response to a rising blood-sugar, frequently the diabetic tissues continue, to a somewhat diminished extent,

to liberate sugar into the blood-stream. It is reasonable to suppose that this failure of the peripheral tissues to remove sugar from the blood as do normal tissues under similar conditions is a demonstration of the inability of the diabetic organism to produce insulin in response to a rising blood-sugar level.

We have shown that the young diabetic is sensitive to insulin, and for this reason it might be expected that the hormone would alter the A.V. difference. Inspection of the curves obtained after glucose and insulin (Figs 2, 3 and 4) shows this to be the case, particularly in certain experiments ; the average curves (Fig. 1B) show a definite, if transitory, action of insulin in this respect, though not in general powerful enough to produce a positive A.V. difference. It appears, therefore, that under the conditions of our experiments, there is a definite but variable effect of exogenous insulin on the A.V. difference of the young diabetic.

The behaviour of the elderly diabetics is of particular interest in view of their resistance to insulin. The fasting A.V. difference corresponded to that of the young subjects, a small negative difference being the rule (Figs 5 and 6). After glucose the majority of the subjects showed a small change in the A.V. difference with the establishment of a positive difference at the 30 min. period, namely, at the time of maximum action of the insulin ; in the remainder the initial negative difference persisted, and in some cases increased in magnitude. Such positive differences tended to disappear after a time, with re-establishment of an excessive venous blood-sugar. In the case of the elderly diabetics, therefore, there is usually a slight and transitory increase in the A.V. difference after glucose (Fig. 1c). This is an interesting observation and suggests that in these subjects, despite their initial high level of blood-sugar, ingestion of glucose in some way increases insulin activity at the periphery. The response of the A.V. difference to glucose was little altered by the simultaneous injection of insulin, the average curves (Fig. 1c) showing no change ; this finding suggests that in these patients the peripheral action of insulin is enfeebled. We emphasise, however, that there is, on the average, very little difference between the peripheral effects of insulin in the sensitive and resistant cases : that in both these groups these effects are considerably less in evidence than in the normal subjects ; and that in contrast, the difference in the response of the arterial blood-sugar to insulin in the two groups is striking (Fig. 1).

The nature of the reducing substance in arterial and venous blood. In order to determine if a substance other than glucose is responsible for the reducing power of venous blood, when this is higher than the corresponding arterial blood, sugar estimations were carried out before and after treatment of the blood filtrates with yeast as described by Peters and van Slyke (35). Using MacLean's method it was found that all but a minute fraction of the reducing substances in blood are removed by yeast, and that there is no significant difference between venous and arterial blood in this respect.

Discussion.

The A.V. difference. Since Foster (13) introduced the simultaneous determination of the sugar content of arterial and venous blood as a means of studying the rate of removal of sugar from the blood by the tissues, numerous workers have used this method for investigating the peripheral utilisation of sugar under various conditions. In our work we have frequently obtained values for the sugar of venous blood which exceeded those of the corresponding arterial blood by more than the experimental error involved in the determination. It is interesting to note that while such negative differences were rare in normal subjects they frequently occurred in diabetic individuals : in normals 2% only of the total determinations were negative, whereas in both types of diabetic negative differences constituted 60% of the total. A perusal of past records reveals that negative values, often of considerable magnitude, have been obtained by a number of investigators, in normal subjects after insulin and adrenaline (17 and 18), in diabetics (4, 18, 28, 29 and 36), and in the depancreatised animal (14 and 19). Negative differences have been observed in the fasting normal subject (15 and 18), but apart from this there is hardly any reference to their occurrence under strictly physiological conditions.

That at certain times the venous blood is found to contain more reducing substance than the corresponding arterial blood has as yet no satisfactory explanation. We are satisfied that the production of stasis for a short period before withdrawal of venous blood is without effect upon the blood-sugar. We have shown, also, that the reducing substance is removed by yeast. The frequency of negative values in the diabetic and their rarity in the normal subject, although exactly the same technique was used in all experiments, seems definitely to exclude an accidental origin, and we conclude that the difference is a real one.

When we try to explain these negative A.V. differences we are at once on difficult ground. The excess venous sugar might be derived either from the glycogen of the tissues, or from the free sugar stored in them ; a more remote possibility is that the sugar is formed from some non-reducing precursor in the arterial blood during its passage through the tissues. It can be said at once that it is most unlikely that the source of the sugar is the glycogen of the muscles. We know from the experiments of Mann and Magath (33) on the hepatectomised animal that even under conditions of extreme need the muscles are unable to yield their glycogen to the blood as sugar. We must, therefore, turn our attention to the free sugar of the tissues. Lawrence (29) suggests that because, in his experience, negative A.V. differences occur at a time when the diabetic subject is uninfluenced by insulin, this hormone causes the tissues to store more sugar than they can retain when its action ceases. Bose (4), whilst remarking the failure of many investigators to comment on the negative differences which appear in their work, is inclined to agree with Lawrence.

Although we are not in a position to bring fresh evidence we emphasise that the negative A.V. differences are characteristic of the diabetic state and consider this fact to be very important. It is at least possible that these abnormal differences are in some way related to the presence of excessive quantities of *free* sugar in the tissues, and that this sugar accumulates because of some impairment of the action of, or deficiency in, insulin, which under normal conditions promotes the *utilisation* of sugar, either by oxidation or by conversion to glycogen. Especially when the blood-sugar is high, the tissues are able rapidly to absorb sugar from the blood and to retain it, at least temporarily, as free sugar ; indeed, the avidity of the normal tissues for sugar is remarkable. Folin and his co-workers (12) found that within 5 min. of injecting 2 g. of glucose per kg. body-weight into a nephrectomised dog about 70-80% of the injected sugar had passed from the blood to the tissues, and that after a lapse of 20 min. the muscle sugar had increased by about 100 mg.% ; by this time the muscle glycogen had scarcely altered. Furthermore, these workers found that when the blood-sugar level was rapidly raised to 700 mg. % there was an immediate and rapid transference of sugar to the skin, in which the concentration often rose to 300-500 mg. %. In the case of the skin particularly, but also in regard to the muscle, as the blood-sugar fell the process was reversed, and sugar diffused back into the blood-stream. The high blood-sugar common in the diabetic would favour the accumulation of sugar in the tissues, and because of the relative weakness of insulin action a large proportion of this sugar might remain unchanged. It seems possible, therefore, that the excess sugar in the venous blood of the diabetic is derived from sugar which at some earlier time has been taken up and retained as such by the tissues and skin during a hyperglycæmic phase. This simple explanation is not, however, in harmony with the experimental findings. Firstly, one would expect that after a fasting period of 12-15 hours equilibration of the tissue–and blood-sugar would be complete, whereas negative differences frequently occur at this time ; secondly, if diffusion alone is responsible, negative differences should be confined to times when the blood-sugar is falling, but this is not the case. The existence of a negative difference coincident with a rising blood-sugar is, at the moment, inexplicable. It seems to us unlikely that changes in the rate of blood-flow, or in the degree of hydration of the blood, enter into the problem. Marks (34) has shown that extracts of the anterior pituitary diminish glycogen formation in the muscles under the action of insulin ; the possibility that this is effected by a transformation of muscle glycogen to sugar, though it seems unlikely, deserves investigation.

The nature of insulin resistance. It has been shown that according to their response to the insulin-glucose test diabetics fall into two groups, namely, the insulin-sensitive, and the insulin-resistant. In our work we were not directly concerned with the possibility suggested by Himsworth that diabetics might be typed according to their response. It happened that the young patients were uniformly sensitive, and the elderly patients resistant,

but we hesitate to conclude that this is always the case; the possibility of a differentiation on the basis of the insulin-glucose test is being more fully investigated. Our interest centered on the arterio-venous blood-sugar difference, and on the mechanism of the resistance exhibited by certain diabetics.

The metabolic changes which occur in the normal subject as the result of the simultaneous administration of glucose and insulin are identical with those which are associated with the fall of the blood-sugar to normal levels after ingestion of glucose, and which must be ascribed to the presence of endogenous insulin. In all diabetics these signs of the production of insulin are largely lacking. In the case of the sensitive diabetic the pronounced effect of injected insulin shows quite clearly that their defective metabolic response to glucose is due to a deficiency of insulin, a view which is in accordance with that of Falta (11), Himsworth (21) and MacBryde (32). On the other hand, the defect in the resistant diabetic is probably not directly the result of a lack of insulin because, as we have shown, when insulin is injected into the blood-stream its metabolic action is but feebly exerted. We may assume, therefore, that any endogenous insulin that might be produced in these subjects would likewise be unable to exert its normal action on the metabolism of carbohydrate. The resistance to insulin which is manifested in these patients must be due to some extra factor which acts antagonistically towards insulin; it is at least possible that their symptoms are largely, if not entirely, due to this anti-insular influence.

At which point in the process of carbohydrate metabolism is this anti-insular effect exerted? Under fasting conditions the sugar in the blood is in process of transit from the main storehouse, the liver, to the places of consumption, of which the chief is muscle. At such times its concentration in the blood represents a dynamic equilibrium, slight displacement in one direction or the other being immediately compensated by fine adjusting mechanisms at present little understood. Gross disturbances of the blood-sugar level, such as occur after ingestion of carbohydrate, or during the rapid utilisation of carbohydrate in the muscles during exercise, are corrected by more powerful agencies, the pancreas on the one hand and the thyroid-adrenal system—possibly under the control of the pituitary—on the other. As regards the pancreas we know that in response to the increase in blood-sugar which occurs during absorption of carbohydrate, insulin is secreted; that this insulin acts upon the liver, tending to stem the flow of sugar from that organ, and that it also acts upon the peripheral tissues, causing them to abstract more sugar from the blood passing through them; by these means a gross increase in the concentration of sugar in the circulating blood is prevented, and the normal fasting level is in time restored. Thus it will be obvious that two distinct spheres of insulin activity exist: the central, or hepatic, sphere; and the peripheral sphere. It follows that if in a given subject insulin fails, either entirely or partially, to prevent hyperglycæmia during absorption of glucose it must be because it is prevented from exerting its effect at either, or both, these sites of sugar utilisation.

In the normal subject the peripheral action of insulin is clearly shown by the increase in the A.V. difference which occurs after ingestion of sugar, and by the fact that injection of insulin increases this difference. In view of the magnitude of the A.V. difference during the action of insulin, and the large mass of muscle in the body, it is probable that the removal of sugar by the peripheral tissues is mainly responsible for the prevention of excessive hyperglycæmia during the early stages of absorption of carbohydrate from the gut. As soon, however, as the tissues are replete with sugar, the hepatic storage mechanism must be called into action to control the blood-sugar level; evidence for this is afforded by the diminished hyperglycæmia which follows a second dose of sugar given after a short interval. In sensitive and, to a large extent, in resistant diabetics in our experience the A.V. difference is not increased after sugar ; partly for this reason, and partly because the passage of the absorbed sugar through the liver continues unchecked, the blood-sugar reaches high levels and a large proportion of the ingested sugar is excreted in the urine. There is, therefore, in diabetes a partial breakdown of the peripheral mechanism for dealing with sugar, and in addition the central storage of sugar is impaired. In the case of the sensitive diabetic we have found that injected insulin causes the peripheral tissues to retain more of the sugar contained in the blood passing through them, but because the extent of this effect is less than in the normal with the same dose of insulin, whereas the absolute diminution of the hyperglycæmia is greater, it appears highly probable that the hepatic mechanism is chiefly influenced, with the result that less of the sugar absorbed from the gut escapes through the liver into the general circulation under the action of insulin, a view which is in agreement with the experimental work of Cori and Cori and Goltz (7). In other words, in the sensitive diabetic, as in the normal subject, insulin promotes storage of sugar in the liver.

On the question of insulin resistence we do not find ourselves in complete agreement with the conclusion of Himsworth (21) that the principal site of failure of insulin action is in the peripheral tissues. There is evidence of peripheral utilisation of sugar in the resistant patients after ingestion of glucose, and in general this peripheral activity is little affected by injection of insulin. In this sense it might appear that these patients are resistant to the peripheral action of insulin, but it can be no more than a partial explanation of their resistance. In comparison with the normal both sensitive and resistant cases show, in general, a smaller peripheral response to insulin. When, however, the peripheral effects of insulin in the two types are compared, the difference is insufficiently great to warrant the conclusion that the comparative absence of change in the arterial blood-sugar curve of the resistant diabetic after insulin is due entirely to impairment of the peripheral action of insulin. It appears to us more probable that the resistance exhibited by these patients is mainly due to some weakening of the normal influence of insulin in promoting storage of sugar in the liver.

If insulin resistance is indeed a hepatic phenomenon a further similarity between the resistant diabetic and the animal receiving extracts of the anterior pituitary is established. According to Houssay (23) the anterior pituitary does not exert its diabetogenic influence in the toad in the absence of the liver, but this is perhaps not surprising in view of the severe disability of the hepatectomised animal. The role of the liver in insulin sensitivity is emphasised by the work of Cope and Marks (6) who were able to show that administration of pituitary extracts to an animal increases its sensitivity to the hyperglycæmic action of adrenaline; and that in the absence of the pituitary adrenaline is unable to raise the blood-sugar from hypoglycæmic levels. This synergism between the pituitary and adrenaline is also apparent in the work of Képinov (26). It would seem, therefore, that the pituitary is able to oppose the action of insulin by rendering the liver glycogen more susceptible to the mobilising action of adrenaline; this was termed by Young (40) the glycotropic action of the pituitary. While it is doubtful whether this glycotropic stimulus could ever be called into play at the normal level of blood-sugar, it is even more unlikely to occur at the hyperglycæmic levels of the diabetic, unless, as Falta (11) has suggested, the threshold of the stimulus is abnormal in the resistant diabetic. For this reason it is necessary to postulate the existence of another pituitary factor, the glycogenolytic factor (Young), which causes an intense release of sugar from the liver, and which is supposed to be responsible for the hyperglycæmia which results from the continued injection of anterior pituitary extracts into animals, an effect which is not necessarily an accompaniment of the glycotropic, or adrenaline sensitising, action of these extracts. It is possible that this glycogenolytic effect leads in turn to an increased rate of gluconeogenesis. Thus two modes of liberation of sugar from the liver are implied, in both of which the pituitary exerts its influence: adrenaline (sympathetic) release, an emergency reflex mechanism synergised by the glycotropic factor; and the continuous supply of sugar to the blood to compensate for the metabolic consumption of carbohydrate, a process which is augmented by the glycogenolytic factor. Our observations on the human diabetic suggest that disorders of carbohydrate metabolism may involve two such factors. While from our previous experiments (9) we formed the opinion that the change in the insulin response of the rabbit is due to the presence of the adrenaline-sensitising factor in the positive sera, the present work suggests that the failure of insulin to control the rise in blood-sugar resulting from ingestion of glucose in certain diabetics is due to the presence of an excess of a glycogenolytic factor acting upon the liver. In this connection it is of interest to note that Képinov and Petit-Dutaillis (27) claim to have demonstrated the presence in the blood of the diabetic animal of a substance capable of raising the blood-sugar of a recipient animal rendered sensitive by a pancreatic graft.

Some interesting experiments carried out by Boller and his co-workers (3) have a possible bearing on our work. These authors found that, whereas

the presence of injected insulin in the blood of sensitive diabetics could be demonstrated by transfusing some of the blood into an insulin-sensitive recipient, this was not possible with insulin injected into the resistant diabetic. They suggested that the blood of the insulin-resistant diabetic might contain a substance which antagonises the activity of insulin. The detection in human blood of a substance capable of raising the fasting blood-sugar level of an animal has not, so far as we are aware, been reported. In order to raise the fasting blood-sugar of animals with pituitary preparations a course of injections over periods of several days or months is necessary ; the effect develops only slowly and a single injection of blood-serum cannot therefore be expected to yield a positive response.

SUMMARY AND CONCLUSIONS.

The effect of an intravenous injection of insulin on the rise in arterial blood-sugar following ingestion of glucose was studied in a number of normal and diabetic subjects. Some diabetics are sensitive to insulin, which either diminishes or abolishes the rise in arterial blood-sugar as in the normal ; others exhibit considerable resistance to the hormone so that the arterial hyperglycæmia is little affected.

A study of the arterio-venous blood-sugar difference revealed that in spite of the disparity in the effects of insulin on the arterial blood-sugar in these patients there is no corresponding difference in the peripheral action of the hormone, which in both types is less than it is in the normal subject. For this reason we conclude that although in all diabetics there is an enfeeblement of the peripheral action of insulin, resistance to insulin as determined by the insulin-glucose test is mainly due to the inability of the hormone to exert to the full its control over the storage of sugar in the liver.

The frequent occurrence of negative A.V. differences (venous sugar exceeding arterial) in the diabetic, and their rarity in the normal subject, is discussed. At the moment no completely satisfactory explanation of these negative differences is possible.

In the present state of our knowledge it appears that the insulin-sensitive diabetic is suffering from a lack of insulin, and that the symptoms of the insulin-resistant patient are, at least in part, due to the activity of some factor which opposes the action of insulin on the storage of sugar in the liver. The possible relation between the relative inability of insulin to control hepatic glycogenolysis in the resistant patient and pituitary dysfunction is discussed.

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THE EFFECT OF DIFFERENT DIETS ON THE INSULIN
SENSITIVITY OF DIABETICS.*

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THE use of high carbohydrate diets in the treatment of diabetes dates back to the last century at least. In 1871, Donkin (7) was undoubtedly obtaining beneficial results in many cases by employing his " skimmed milk cure." This consisted in giving nothing but skimmed milk daily up to 7 pints. Assuming the ordinary composition of skimmed milk this is equivalent to a 1,550 calorie diet with 214 g. of carbohydrate, 142 g. of protein and 13 g. of fat. The improvement he obtained in many cases was undeniable and was measured by observations of the urinary specific gravity, urinary volume, qualitative tests for sugar and gain in weight. Up to the end of the pre-insulin era this diet was periodically in vogue. We find the same type of diet hiding under many different aliases. Thus von Noorden (22) proposed the " oatmeal cure." In favourable cases the glycosuria rose initially but afterwards sank or disappeared after one or more oatmeal periods. In 1911, Klemperer (20) introduced periods of glucose feeding to his diabetics and was similarly able to show lessening of the glycosuria as a result of this feeding. This method has been rediscovered by Gibson (12) who reports that periods of feeding on sugar accompanied by increased insulin produce increased tolerance and that, as a result, in certain young subjects dietary management alone without insulin may control the diabetes. There does, therefore, appear general agreement over a period of some 50 years, that amelioration of diabetes may occur after periods of feeding on a high carbohydrate diet. The results recur so constantly in past records that they cannot be disregarded. They indicate that the diabetic patient's response to carbohydrate is in some way modified by his recent dietetic experience.

No valid explanation of this paradox has been put forward.

In 1919, Hamman and Hirschman (14) carried out experiments on normal subjects which have some bearing on this phenomenon. These patients were given repeated doses of glucose and the resulting blood sugar curves plotted. They found that the response of the blood sugar to the second

dose of glucose was less marked than the first and that a flatter sugar tolerance curve resulted. The phenomenon was later studied by Staub (27) and Traugott and usually goes under the name of the Staub-Traugott effect. Hamman and Hirschman noted a similar but slighter effect in diabetics and suggested that there was no qualitative but only a quantitative difference between the carbohydrate metabolism in normals and diabetics in this respect. This response to repeated doses of sugar produces such progressively lowered sugar tolerance curves that, in a renal diabetic, Gibson and Larimer (13) were able to provoke hypoglycæmic symptoms by such means.

In 1927, Sweeney (28), showed that glucose tolerance curves were materially modified by the previous diet. His subjects were young healthy males and the diet was adhered to for two days before the test. Those on a diet rich in carbohydrate showed a marked increase in sugar tolerance as measured by a flatter curve compared with patients previously on a fat or starvation diet. Hynd and Rotter (18), in 1931, attempted to show variations in sensitivity towards insulin of animals on different diets. They showed that laboratory animals fed on a carbohydrate free diet though showing no change in muscle and diminished liver glycogen are less sensitive to insulin injections than those fed on a carbohydrate rich diet. It was easier to convulse animals who had been on a carbohydrate rich diet than those who had been on a carbohydrate free diet. This work has been recently followed by a series of papers by Himsworth (15, 16, 17) in which he confirmed the fact that in healthy human subjects changes in the composition of the diet altered the tolerance to sugar as measured by an ordinary blood sugar curve and also tested the reaction to insulin as measured by the fall of blood sugar after a standard injection of insulin. He attempted to correlate the two phenomena and was able to express the correlation by a straight line. This indicated that any increase in sugar tolerance prompted by a diet high in carbohydrate was entirely accounted for by the increased insulin efficacy obtained. The " sensitisation " of insulin he suggested was due to a hypothetical activating substance secreted by the liver.

If this hypothesis is correct it has an obvious bearing on the working of higher carbohydrate diets in diabetics. It is an established clinical fact that certain diabetics if changed to an equicaloric diet but containing raised amounts of carbohydrate do not require any increase in their insulin dosage. The papers of Ellis (8), Richardson (24), and Ercklentz (9) among many others document this observation. Can we explain this undoubtedly increased tolerance on the supposition that their insulin is in some way more active and effective ? The following experiments were carried out to test this theory ; while they were in progress some work by MacBryde (21) appeared on similar lines.

MacBryde measured the response to subcutaneous and intravenous insulin of patients on different diets. On the basis of their response to insulin he divided them into insulin sensitive and insulin insensitive groups. He found that the former failed to gain tolerance on higher carbohydrate diets

while the resistant cases were invariably able to do so. This gain was accompanied by an increase of insulin sensitivity in certain cases. He measured carbohydrate tolerance by glucose tolerance curves done from fasting blood sugar levels and then measured the area enclosed or the percentage rise in blood sugar. Results from these two measurements were widely discordant in some cases and it is not clear that the sugar tolerance curves were carried out from the same blood sugar levels and are therefore comparable. The conclusion cannot be drawn from his work that there is an invariable association between changes in sugar tolerance and in the response to insulin. His paper serves to emphasise the great difficulty in investigating this subject and the contradictory results obtained.

Methods.

Untreated diabetics were put on a diet and a dose of insulin so adjusted that a small amount of sugar was excreted. When the sugar excretion was stable an insulin depression curve was obtained. They were then transferred to an equicaloric diet containing larger amounts of carbohydrate, the dose of insulin remaining constant. In favourable cases the sugar excretion was only slightly increased and when this was stable a further insulin depression curve was obtained. By comparing the areas of these two curves any change in the power of insulin to lower the blood sugar on the two diets was · elicited.

The first essential was to obtain patients who showed increased sugar tolerance as measured by their ability to utilise nearly all the added carbohydrate since only then could any useful attempt be made to compare the areas of their insulin depression curves. If a patient was unable to tolerate the added carbohydrate the experiment was not proceeded with, since there was no point in comparing the effectiveness of insulin in such a case apart from the added difficulty that the noon blood sugar was naturally much higher and direct comparison of the curves therefore unwarranted. The results of Collens and Grayzel (17), who investigated the response of diabetics to intravenous insulin, indicate that the percentage drop in blood sugar after a standard dose of insulin is proportional to the initial height of the blood sugar. This necessary limitation naturally excluded many patients and in particular it was found impossible to increase the diet in obese diabetics without nearly all the added sugar being excreted. This is in accord with the experience of Richardson though directly opposed to the experience of MacBryde. Other cases which had to be excluded were those in which any infection intervened, in one case a severe cold and in another a middle ear infection, since this inevitably produces a variable disturbance on insulin action.

Only previously untreated diabetics were used for experimental purposes. They were given an initial diet of about 30 calories per kilogram with protein to the extent of 1½ g. per kilo. In the first period an actual carbohydrate content of usually 100 g. was given. In the second dietetic period the calorie

value of the diet was kept constant, the protein values remaining unchanged but the carbohydrate fraction of the diet was raised by 100 grams and the fat fraction correspondingly reduced. It was in diets with carbohydrate values between 100 and 200 g. that Himsworth (17) found the greatest changes in insulin sensitivity in the normal. The carbohydrate was apportioned throughout the four meals in the proportions of one-third, one-sixth, one-sixth, and one-third. It was not possible to continue the experiment by a further alteration of the diet since the patients were unwilling to remain indefinitely in hospital. Their usual stay was a month. The diets were carefully checked by the Dietetic Sister for any wastage. Since exercise produces profound changes in the blood sugar, the patients were either kept at rest for the whole of the experimental period or were allowed up for two hours in the evening and walked round the ward. No change in the amount of the exercise was allowed during the experimental period. It was not thought possible to evaluate carbohydrate tolerance by employing the ordinary sugar tolerance test since this must have been carried out at different blood sugar levels and might have been affected by the height of the renal threshold and subsequent excretion of sugar. The amount of insulin was arranged in the first dietetic period so that when stabilised a few grams of sugar were being excreted daily. The importance of securing this minimal excretion of sugar in order to transfer successfully the patient to the higher diet without obtaining much added glycosuria was not initially realised. The insulin was given in two doses before breakfast and supper. A twenty-four-hourly collection of urine was made and the sugar estimated daily by Bertrand's method. It was found in general that the excretion of sugar remained reasonably stable ; a few patients exhibited wide variations for no apparent reason. In patients who were obviously making no effort to ensure a full collection of urine, the experiment was discontinued. At the end of a period on the initial diet when the insulin had been adjusted until the daily excretion of sugar was 5-10 g. and and been stable for a week an insulin depression curve was obtained. This was done at 12 noon some four hours after breakfast and insulin, when the blood sugar level was relatively constant. Its approximate height was known by previous blood sugar estimations done at this time for some days previously. The patient was either wheeled into a room at the end of the ward or was surrounded by screens to avoid any change due to emotional disturbances. Resting blood samples were taken in the supine position, 5 units of crystalline insulin were given into the brachial vein of the opposite arm and blood sugar samples taken thereafter at two minute intervals for the next twenty minutes. Crystalline insulin was used in order to avoid an initial hyperglycæmia.

The technique of obtaining comparable blood samples at these intervals caused some difficulty, but we finally used a tourniquet on the thumb getting blood from the side or root of the nail with a triangular needle. It was found that if the thumb is kept warm, if the blood is obtained with a deep prick immediately the tourniquet is applied, blood samples corresponded

to the blood obtained from the warm pulp of the thumb. With some
practice 0·1 c.c. of blood can be obtained within 15 seconds of starting to
apply the tourniquet. The crux of the matter lies in keeping the thumb
really warm. The patient should feel warm and be in a warm room, he or
she should have the arm covered from shoulder to wrist either with pyjamas
or bed-jacket, the hand should be kept under the bed clothes the whole
time except when samples are being withdrawn and the hand should lie
on a hot water bottle and be covered by an electric pad. Under these
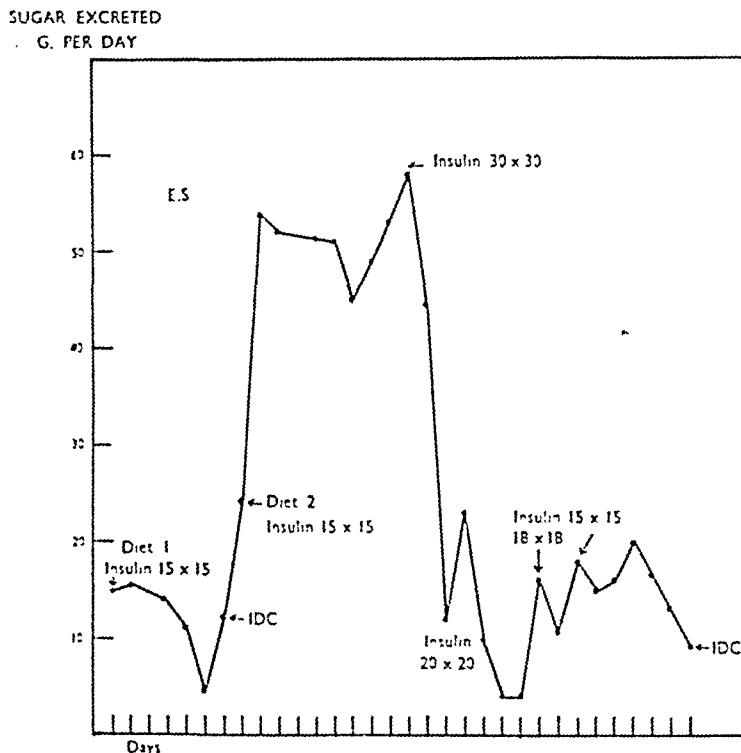
Fig. 1 (Case 2, E.S.).

conditions blood samples corresponded accurately to those obtained from
the warm pulp of the thumb. It is important to adhere strictly to this
routine if consistently reliable results are desired. In two cases, in addition
to the capillary blood samples, venous blood samples were taken. After
the insulin was injected the syringe was left in situ and venous blood with-
drawn after a short time. A further venous sample was taken at the
conclusion of the experiment in the same way. The earlier blood sugars
were estimated by the Folin-Malmros method, using duplicate estimations
on 0·1 c.c. of blood, but later the Hagedorn-Jensen method was used.

As a test of the blood sugar method, a series of blood samples was withdrawn from a normal individual using the technique described. The error of the method was less than 3%. Insulin depression curves done on consecutive days showed an error of 9%.

Fig. 1 shows a specimen graph representing the daily excretion of sugar. By reference to Table I it will be seen that on the first diet with a glucose
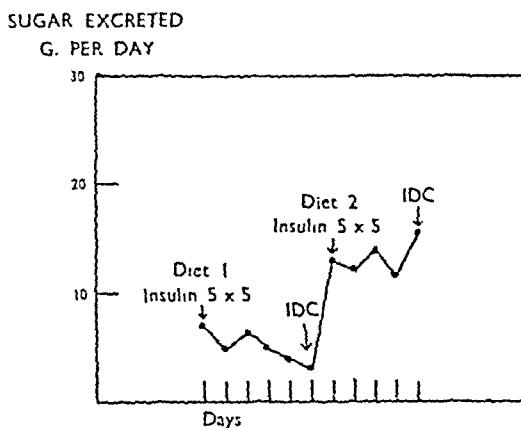
Fig. 2 (Case 1, A.T.).
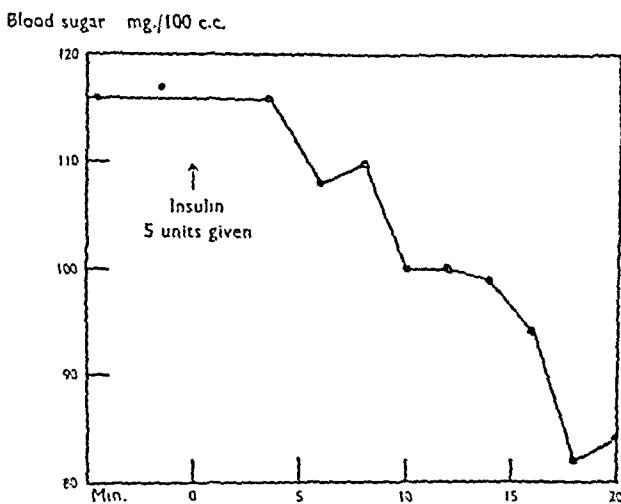
Fig. 3. Insulin depression curve.

equivalent of 179 g. and 30 units of insulin an average of 12 g. sugar were excreted in the days before the insulin depression curve (I.D.C.). The glucose equivalent of the diets is calculated from the usual assumption that all of the carbohydrate, 10% of the fat and 60% of the protein are convertible into glucose and are in fact so converted. After the change over to the higher diet the sugar excretion rises abruptly and the insulin had to be temporarily increased to 60 units a day. It was then reduced to 40 units, 36 units and finally 30 units a day. Some patients were able to take the increased diet

without this temporary increase of insulin, (Case A.T., Fig. 2) in others the insulin had to be increased initially and then slowly reduced. In a similar type of experiment, Ellis was able to reduce the insulin dosage from 192 units to 9 units a day in one case. The sugar excretion on the second diet with the same insulin was 18·5 gms. a day. The patient therefore showed an apparent increase in carbohydrate tolerance since she was able to utilise nearly all the added carbohydrate.

At the points marked I.D.C. insulin depression curves were obtained in the way described. These are shown on Figs 3 and 4. By referring to the column marked I.D.C. (Table I) the area described below the resting level is given in milligram minutes. This area was measured by redrawing
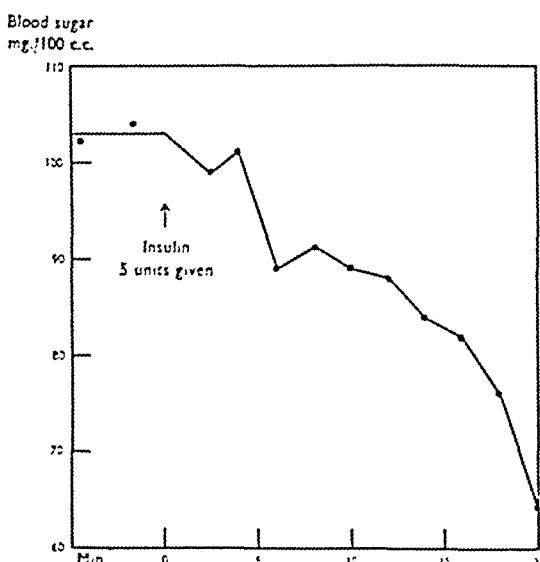
Fig. 4.   Insulin depression curve.

the curve on a large scale and using a planimeter. It is arbitrarily limited by the line drawn at 20 minutes. This figure is preceded by a number denoting the height of the blood sugar levels from which the curve starts. In this case the levels differ by 13 mgms. and the resulting areas should be comparable. It will be seen from the last column there is an increase in the area of the I.D.C. of 24 mg. minutes. This increase of 8% is within the limits of error of the method, and in this case no increase in insulin efficacy has been demonstrated. The results of other patients are similarly tabulated.

Two patients (F.W. and G.G.) showed no change in the I.D.C. areas on the two diets. In the others increase or decrease of the I.D.C. areas took place. The results were averaged by adding together all the initial areas and all the final areas. These come to 2,990 mg. minutes and 3,022 mg. minutes respectively ; an insignificant increase of 1%. All the cases on

glycogen stores in the liver, the less glycogen the easier were convulsions obtained. Himsworth found on normal subjects that the depression curve was steeper and more pronounced in those fed on a carbohydrate rich diet. It is possible that the different results may be explained by the different types of animals used, namely, omnivora, carnivora, herbivora, though Himsworth's work on human subjects appears conclusive.

Again it is not clear how much this improved effect of insulin in animals on different diets is due to increased peripheral utilisation or to diminished sugar production in the liver. An experiment of Soskin's (26) may throw some light on this question. He measured the fall of blood sugar in dogs after evisceration which had previously been fed normally or starved for three weeks. He obtained identical curves in both groups of animals. Now we know from numerous experiments on " hunger diabetes " (6) that the starved animal would have given a diabetic curve after glucose and presumably a diminished fall of blood sugar after a dose of insulin as compared with the normally fed group. The differences in the insulin depression curve would then have been due to the action of insulin on the liver since peripheral storage took place at the same rate in the two groups. If one could assume that insulin is relatively more effective in inhibiting glycogenolysis than in inhibiting gluconeogenesis the paradoxical effect of insulin on different diets might be explained. A greater fall of blood sugar would then be produced in those animals with large amounts of glycogen in the liver and a smaller fall in those animals with less glycogen stored but a rapid rate of gluconeogenesis. In the case of the diabetic who is only under the influence of insulin for some 12 hours out of the 24 it may be that on a higher diet of carbohydrate increased storage of glycogen does not occur. If this is the case insulin depression curves would not show any alteration. This explanation is admittedly highly speculative, but in view of Soskin's work it seems that in the past too little attention has been paid to the role played by the liver.

SUMMARY.

1. Previously untreated diabetics were observed on two equicaloric dietetic periods, the second containing an increased amount of carbohydrate.

2. Their daily urinary excretion of sugar was measured as an indication of their sugar tolerance.

3. At the end of each dietetic period an insulin depression curve was obtained after the intravenous injection of 5 units of insulin and its area measured.

4. In 9 unselected cases no significant difference was found between the sum of the areas of the first insulin depression curves and the sum of the

areas of the second, though all these patients could utilise most of the added carbohydrate.

5. It is concluded that the capacity to utilise increased carbohydrate shown by certain diabetics when transferred to a higher diet bears no constant relation to the phenomena studied in the insulin depression curve.

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It would seem that unless skin and bone blood flow vary greatly and within wide limits, the upper part of the forearm offers a suitable preparation for the plethysmographic study of muscle blood flow in the human subject. We shall here deal only briefly with some of our observations on blood flow in active muscle and confine our attention to other aspects of the limb circulation.

In the first place we find that Hewlett and Zwaluwenburg's (8) method is not always suitable for measuring blood flow in patients in whom the main arteries of the limb are blocked. The method consists essentially in measuring plethysmographically the rate of swelling of the limb when pneumatic pressure is applied above the plethysmograph to a degree sufficient to arrest for a time the venous outflow, but insufficient (less than diastolic pressure) to obstruct arterial inflow. The pressure commonly used lies between 50 and 70 mm. Hg. It is clear, however, that when the main arteries are blocked and circulation is maintained through smaller collateral vessels, the pressure in which cannot be estimated, the application of these or lower pneumatic pressures over the collateral vessels may compress them and materially reduce or even arrest arterial inflow. For example, in one patient in whom the subclavian artery had been tied, the application of a pressure of 50 mm. on the upper arm arrested blood flow to the forearm. Inflow curves were obtained by using lower congesting pressures. The blood flows estimated from these were so much smaller than those indicated by skin colour and temperature, that we believe even the low congesting pressures used reduced arterial inflow.

Secondly, we find that in normal subjects, the blood flow measured in this way does not represent only the arterial inflow to the portion of the limb enclosed within the cuff plethysmograph, as Lewis and Grant (12) assumed. The venous filling of the enclosed portion of the forearm may be considerably influenced by the venous return from the part lying distally to the plethysmograph. To ensure that the inflow curves measured in this way represent arterial inflow to the enclosed part of the limb alone, the distal circulation requires to be arrested by a sphygmomanometer cuff applied to the forearm immediately below the plethysmograph. In some circumstances there is but little difference between the blood flows measured when the distal circulation is free and when it is arrested; in others the difference is stiking, the blood flow being much less when the distal cuff is inflated. We have not found it necessary to define precisely the factors controlling the difference, but the chief appears to be the rates of filling of the venous reservoirs in the two parts of the arm when the congesting pressure is applied. It seems that if the distal veins fill more rapidly than those enclosed within the plethysmograph then blood passes from them into the forearm veins, thus adding to the rate of swelling in the enclosed portion of the limb. In practice, a considerable reduction in the forearm blood flow by arresting the distal circulation indicates a greater blood flow in the distal than in the enclosed portion of the limb. The difference between the

blood flows, however, can only be used as an index and not as a measure of the distal blood flow. An example of the effect of arresting the distal circulation is given in Fig. 1, which shows the forearm inflow curves in a resting normal subject when cold (left hand curves) and when warm (right hand curves). The upper curves were inscribed with the distal circulation free, the lower ones with this arrested. In the cold subject the slope of the inflow curve is but slightly reduced, while in the warm subject the reduction is to about one third of the previous angle. (See also Fig. 7, page 132, for corresponding blood flows). Separate observations on the hand and forearm show that in the cold subject the blood flow is no greater in the hand than in the forearm,
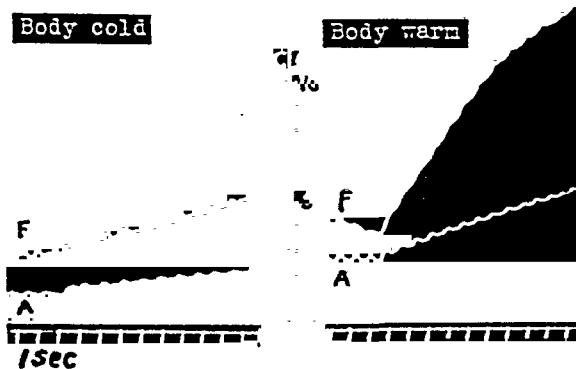
Fig. 1. Forearm inflow curves; left hand curves with body cold; right hand curves with body hot. Upper curves with distal circulation free, lower curves with distal circulation arrested. D.S. 2.3.37; limb volume 500 c.c.; bath temperature 30°; room temperature 11·5°. Time marked in seconds.

while in the warm subject, the hand blood flow greatly surpasses that in the more proximal part of the limb.

Further observations made in this way have revealed important differences between the circulation in the forearm and that in the hand. Before discussing these a word is required about plethysmographic methods.

Method.

The details of the cuff plethysmograph and of the method of measuring the blood flow have already been fully described (S. 12) and it is unnecessary here to do more than indicate the main points of these and the modifications now required. As originally devised the plethysmograph was a truncated cone of aluminium, the two ends of which were closed by fitting over each rubber tyre tubing and inserting the arm through the holes. This form is suitable only when the arm is at rest. When the muscles are contracted

leaks are liable to develop between the rubber and the skin at the grooves between the muscles. We have therefore modified the plethysmograph by placing inside the cone a sleeve of thin rubber which fits the arm loosely.* The ends of this sleeve are everted and fixed over the ends of the metal cone. To prevent folds of this sleeve from blocking the mouths of the outlet tubes of the plethysmograph (which happens occasionally) an inner cone of perforated metal sheeting is inserted between the outer cone and the rubber sleeve. To prevent the sleeve from bulging at the ends it is here supported by stout rubber sheeting fixed to the plethysmograph over the everted ends of the sleeve and through which holes are cut to the shape of the forearm. The records obtained with this modified plethysmograph do not differ from those of the original; it does not leak and in spite of vigorous and repeated contractions of the forearm muscles does not alter its position on the arm. The other details remain as previously described. When the plethysmograph is in position on the arm it is filled with water under pressure to ensure that all air is removed and that the sleeve is closely applied to the arm and to the rubber diaphragms at the ends. A finger is inserted under these diaphragms to let out any air beneath them. The pressure is then released and water withdrawn until only a short column remains in the vertical outlet tube. This wide tube is connected to a float recorder with a vertical writing lever, a device which greatly facilitates the estimation of blood flow from the inflow curves; the recorder usually employed gives an excursion of 4 to 5 mm. to 1 c.c. change in the plethysmograph. A sphygmomanometer cuff is applied to the lower forearm immediately below the plethysmograph. This cuff can be inflated from a reservoir (usually at a pressure of 200 mm. Hg) when it is desired to arrest the circulation to the hand and lower forearm. To obstruct the veins for measuring blood flow, a second cuff is applied to the upper arm. This can be rapidly inflated at will from a reservoir maintained at a pressure of 50 to 70 mm. Hg. Like Lewis and Grant (12) we have observed the precaution before beginning observation of resting the subject for at least half an hour with the plethysmograph in position, the cuffs applied and the arm in a water bath maintained at a desired temperature, usually 30°C. Prolonged rest is necessary because of the long lasting nature of the vasodilatation provoked by previous use of the arm. It is important to have the subject as comfortable and relaxed as possible; tension on the muscles of the forearm or movements of other parts of the body may materially alter the blood flow through the arm under observation. It is usually unsatisfactory for this reason to prolong observations beyond two hours, since after that time the subject becomes uncomfortable and restless. We chose 30°C for the arm bath as a temperature that neither constricts nor dilates the limb vessels unduly and is not far removed from the usual surface temperature of the exposed forearm. Previous observations on the fingers and hand showed us that at this temperature reflex changes of volume are

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* Krogh, Landis and Turner (9) have used a similar modification.

well displayed, lower temperatures tending to interfere with dilatation and higher with constriction. We have also controlled the state of the cutaneous vessels by keeping the subject comfortably warm, cold or hot according to the circumstances. We shall see, however, that whether the subject is hot or cold makes but little difference to forearm blood flow. The forearm has been slightly dependant, on a level with the xiphisternum ; the total pressure on the arm veins due to gravity and the plethysmograph has been no more and usually less than 10 mm. Hg. No advantage is gained by raising the arm above the heart level.

Arrest of the circulation to the hand and lower forearm can be maintained without material discomfort for 30 to 45 minutes but it is not usually necessary so to maintain it. There is no difference between the resting blood flows recorded at intervals over a period of half an hour when the circulation to the hand is maintained arrested and those when the distal circulation is arrested intermittently, while the inflow curves are inscribed. It is our rule to allow an interval of 10 to 15 seconds to elapse between the inflation of the lower cuff, to arrest the distal circulation, and the inflation of the upper cuff to inscribe the inflow curve. Inflation of the lower cuff causes a transient increase in forearm volume owing partly to blood being expelled from the lower forearm and partly to the cuff itself pressing against the lower end of the plethysmograph. The increase subsides and the base line is again straight within 10 or 15 seconds.

When the above precautions are observed and the cuff below the plethysmograph is inflated, the base line written by the recorder is usually remarkably level and successive estimates of blood flow agree closely. Such variations of volume and blood flow as occur in the forearm under resting conditions are considerably less than those seen in records from the hand, and specially from the fingers alone. We give the following as an example of successive forearm blood flow rates taken at half minute intervals in a resting subject maintained comfortably warm ; 1·2, 1·0, 1·0, 0·9, 1·2, 1·4, 1·0, 1·4, 1·8, 0·8, 0·9, 1·4, and 1·4 c.c. per 100 c.c. limb volume per minute or an average of 1·2 c.c.. Taken again 15 minutes later the flows were 0·8, 1·0, 1·0, 1·0, 1·1, 0·8, 1·0, 1·1, 1·4 and 1·4, an average of 1·0 c.c.. A point of interest is that these apparently spontaneous variations are as a rule opposite in direction to those occurring in the hand. Simultaneous records from one hand and the other forearm show that when limb volume and blood flow diminish in the hand they usually increase in the forearm. These small variations in blood flow are in chief part vasomotor in origin ; they are greater in some subjects than in others and are almost abolished by sympathectomy. Because of them it is our custom in estimating resting blood flows to take the average of three successive inflow curves. Each curve is inscribed for a period of 10 seconds, with an interval of 5 to 10 seconds between each to allow the curve to return to the original base line.

We have also applied the cuff plethysmograph to the calf of the leg, using a wider metal cylinder to allow of its slipping over the ankle. The occluding armlet is applied immediately beneath the plethysmograph and the congesting armlet above the knee. The knee is partly flexed over the end of the water bath ; the foot and thigh are held in position by sandbags, the muscles being relaxed. The subject lies on pillows, the plethysmograph being at or slightly above heart level.

A number of observations have also been made on the hand and on the fingers alone. For the hand we have used a metal cylinder closed at one end and with a rubber diaphragm at the other, through a hole in which the hand is inserted. For the fingers, separate narrow metal cylinders are applied to one or more fingers which are inserted through rubber diaphragms covering one end of the cylinders ; at their other ends the cylinders are connected to one recorder. The congesting armlet is applied to the wrist. Estimates of blood flows in the hand and specially in the fingers are not very satisfactory when blood flow is rapid ; the inflow curves run straight for only two or three beats before curving horizontally. This happens even with plethysmographic pressure under 10 mm. Hg and with the hand above heart level and seems to be due to the small capacity of the local venous reservoir in relation to the very copious arterial inflow. In the forearm and leg, however, the venous reservoir being apparently so large in relation to the blood flow, except after strenuous exercise of the muscles, the inflow curves run straight for many beats, thus allowing a more accurate estimate of the rate of flow. Under resting conditions, the forearm inflow curve will rise for 2 or 3 minutes, showing a volume increase of 20 to 30 c.c., before curving towards the horizontal. Such long inflow curves show small departures from the straight line, corresponding to the spontaneous variations of blood flow. They are of service in recording the fleeting changes in response to various stimuli which are liable to be missed when shorter successive curves are inscribed.

We have used another method for estimating blood flow in forearm, hand or fingers of a given limb by enclosing the whole limb to the elbow in a long metal cylinder. A sphygmomanometer cuff is applied to the wrist. The fingers and thumb are arranged round a cylindrical rubber support so that they are held parallel and a narrow cuff is wrapped round their bases. The rubber tubes of the finger and wrist cuffs are led outside the plethysmograph and connected to reservoirs from which the cuffs can be inflated at will. The metal plethysmograph is made in three sections to facilitate the introduction of the arm with the cuffs applied. The congesting armlet is applied above the elbow. In this way, records can be obtained from the whole arm and with the circulation to the fingers alone or to the whole hand arrested. The volumes of the parts are measured by displacement ; the blood flows to the fingers, hand and forearm can thus be estimated. The results obtained with this plethysmograph are in good agreement with those made with separate instruments.

Differences between the circulation in the distal and proximal parts of the limb.

Reactive hyperaemia. In view of the effect of the distal portion of the limb on the blood flow to the part enclosed within the plethysmograph we have repeated the observations on reactive hyperæmia made by Lewis and Grant (12),* but find that the values obtained by them require no material modification. The forearm blood flow immediately following the release of circulatory arrest lasting for periods up to 15 minutes is just as great when the distal circulation is arrested as when it is free and our results agree closely with those previously obtained. The only difference is that when the distal circulation is arrested the hyperæmia declines quicker than when it is free, being curtailed by 1 or 2 minutes. This speedier decline in reactive hyperæmia can also be noted in the skin flush. The circulation to both arms is arrested and released simultaneously, while that to one hand is maintained arrested. The forearm flush fades one to two minutes earlier on the side on which the hand circulation is arrested.
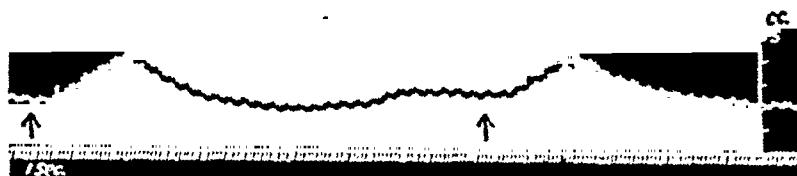
Fig. 2. Forearm volume curve; increase following loud noise (at arrows). D.S. 9.3.37; comfortably warm. Limb volume 690 c.c.; bath temperature 30°; room temperature 13°. Time marked in seconds.

Sensory stimuli. Another difference, with which we shall deal only briefly is the response to sensory stimuli applied to the body. As is known, the general effect of sensory stimuli on plethysmographic records taken from the hand and forearm as a whole, or the hand alone, is a fall in volume and a reduction in blood flow. A few authors (for example, Maragliano and Lusona (15)) have recorded dilator responses as occurring occasionally. In previous and unpublished observations by one of us (R. T. G.) dilator responses were never observed; the constrictor responses were found to be greater in the hand alone than in the hand and forearm together and in the fingers than in the hand. We now find that just as the forearm differs from the hand and fingers in its apparently spontaneous variations so it differs in its responses to sensory stimuli. Such stimuli as a loud noise,

* It may be noted that when Lewis and I were making observations on reactive hyperæmia, it was our custom to use a tightly fitting rubber diaphragm on the lower end of the plethysmograph. — R.T.G.

between 1 and 2 c.c. per 100 c.c. limb volume per minute. Right cervical ganglionectomy was performed for relief of symptoms in the right hand ; sweating was abolished from the arm after operation. Blood flow to the right forearm 19 hours after operation was between 5 and 6 c.c. but declined in the course of a few days and, by 9 days after operation, had fallen to and thereafter remained at about the original level of 1 to 2 c.c.. Fifteen days after operation successive injections of 2 gamma adrenaline raised the blood flow of the left forearm to 10 to 13 c.c. per 100 c.c. limb volume per minute. These observations on the greater dilator response to adrenaline when vascular tone has been regained after sympathectomy in man are in agreement with the earlier work of Dale and Richards (1) on the cat. They show clearly, as Dale and Richards (1) first pointed out (see also Grant (4)) that the factor responsible for the regain of vascular tone following sympathectomy is not circulating adrenaline. Moreover, it is clear that owing to the rapid regain of vascular tone, the finding of a lower blood flow some time after sympathectomy than was obtained by body warming before operation cannot be used as evidence for the existence of vasodilator nerves as it has been by Prinzmetal and Wilson (16).

Change of body temperature. A further difference, and one that we shall consider in detail, lies in the responses to change of body temperature. The large vascular changes that occur in the hands and feet when the body is warmed or cooled are well known. With the forearm alone, however, we find that whether the body is cold or hot, unless it is heated to an unusual degree, makes but little difference to the resting blood flow. We will take first the effects of cooling the body.

(a) *Effects of cooling the body.* In this connection we had in mind Rein and Schneider's (17) observations on the dog, that cooling the body causes not only vasoconstriction in skin but vasodilatation in voluntary muscle. In view of the large proportion of muscle and the low values of the resting blood flow in the human forearm, if any material increase of muscle blood flow occurs on cooling the body this ought to appear as an increase in forearm flow. In the resting subject maintained comfortably warm, forearm blood flow remains at a level of between 1 and 2 c.c. per 100 c.c. limb volume a minute. On numerous occasions we have thoroughly chilled the resting subject by exposing him naked to the air of a cold room at a temperature of 12 to 15°C for periods up to 1½ hours. We do not wish here to deal with the effects of shivering, and usually under these conditions the onset of shivering is long delayed, often not occurring till the subject begins to dress at the end of the observation. Occasional short bouts of shivering may happen, but these are controllable voluntarily. It is of interest to note, however, that uncontrollable shivering may be provoked by a further cold stimulus, as by immersing the feet in cold water or by a draught of cold air on the body. Prolonged cooling in this way is not accompanied by an increase of forearm blood flow. Rather, blood flow falls gradually and slightly, usually to about 0·8 c.c. and on occasion to as

low as 0·5 c.c. per 100 c.c. limb volume per min.. In the hands and fingers under these conditions blood flow also falls to the same low levels. It is clear from these observations that there can be no material, if any, increase in the circulation through the limb muscles when the subject is cooled. For example, in one subject maintained comfortably warm, forearm blood flow was 6 c.c. for an arm volume of 500 c.c. (85% muscle and 9% skin). After cooling blood flow fell to 4 c.c.. Even if we take the unlikely extremes and, neglecting bone circulation, suppose that when the body is warm, muscle blood flow is practically zero, and that when cold the skin blood flow is almost at a standstill, then the maximum possible increase of muscle blood flow, is from practically zero to 4 c.c. for 425 c.c. of muscle, or 0·9 c.c. per 100 c.c. of muscle. Other observations on skin and muscle temperature support this view. It is well known that when the body is cooled the temperature of the fingers falls rapidly towards that of the surrounding air. We find that forearm skin temperature also falls progressively towards room temperature. The rate of fall is much slower than in the fingers, but this can be accounted for by the greater bulk of the part. The rate of fall of the forearm skin temperature in a still room is but little increased by arresting the circulation above the elbow. For example, the difference of the temperature of the two forearms, in one of which the circulation was arrested for half an hour was no more than 0·5°C at the end of the time, room temperature being 15·5°C. We have also measured the temperature of the forearm muscles by inserting into them to a depth of 2 or 3 cm. a needle thermal junction of the type described by Harris and Marvin (7). In the resting arm, muscle temperature lies two to four degrees higher than skin temperature and when the subject is cooled this temperature falls progressively at a rate not much slower than that of the overlying skin. For example, in a resting subject exposed to room air of 15°C, a needle junction, the tip inserted 2 cm. beneath the surface of the dorsum of the forearm, recorded a temperature of 34·1°C ; that of the overlying skin was 32·9°C. Both temperatures fell slowly and steadily. After 75 minutes muscle temperature was 29·4°C and skin temperature 27·4°, a fall of 4·7° and 5·5° respectively. These observations show clearly that in man resting muscle blood flow is very small and that if it is increased at all by cooling the body, the increase is so small as to be negligible. It seems to us probable that the muscle vessels are not affected through the sympathetic nerves by changes in body temperature and that the small decrease of forearm blood flow on cooling the body is due to vasoconstriction in the skin.

(b) *Effects of body warming.* We now come to the effects of warming the body. In these observations, we have usually first cooled the resting subject by exposing him lightly clad to room air for a period of from a half to one hour. In warm weather cooling has been increased by immersing the legs in cold water or by blowing a draught of a fan across the body, or by both. At the end of this time the exposed extremities are cold and the blood flow to the hand, fingers and forearm has fallen to about 1 c.c. or a

little more or less per 100 c.c. limb volume per minute, the average forearm
blood flow rate in eleven observations on three subjects being 1·3 c.c..   The
body is then warmed by covering the trunk with a blanket and immersing
the legs in hot water until the subject feels hot and sweats, and the opposite
exposed hand becomes flushed and hot.   Warming in this way causes a great
increase in blood flow to the fingers and hand, where it rises to between
20 and 30 c.c., and to between 10 and 20 c.c. respectively per 100 c.c. limb
volume, per minute.   In the forearm, as is illustrated in Fig 5, there is but
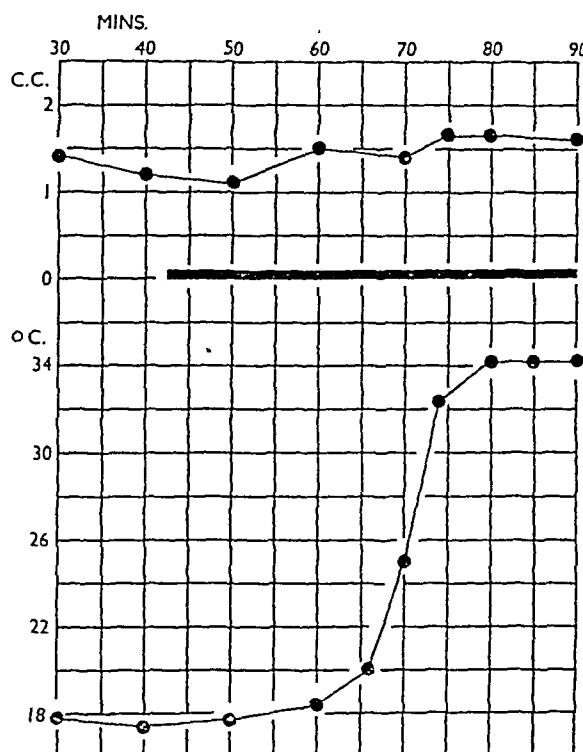
Fig. 5. Effect of body warming on forearm blood flow and on temperature of opposite hand.
R.B.P. 28.10.36.   Right forearm in cuff plethysmograph; forearm volume 600 c.c.; bath
temperature 30°; room temperature 15°.   Upper curve, forearm blood flow measured with
distal circulation arrested and charted in c.c. per 100 c.c. limb volume per min..   Lower
curve, temperature in °C at base of nail, left index finger.   Body warming shown by heavy
line, begun at 42 minutes and continued throughout.

little change, the blood flow rate rising only to between 1·5 and 2·5 c.c.,
the average in the eleven observations already mentioned being 1·7 c.c..
    It is possible to interpret the small increase of forearm blood flow as
due either to vasodilatation confined to the skin or to a larger change in the
skin together with a decrease in muscle circulation.   We have already seen
that any material change in muscle flow is unlikely and further observation
supports the former interpretation.   It is to be remembered that owing
to the small proportion of skin a small increase in forearm blood flow
represents a considerably greater increase in skin flow.   In the example

illustrated in Fig. 5, the blood flow rose from 6·7 c.c. to 11·0 c.c. for a forearm volume of 600 c.c., skin being 11% of the volume. If the increase is due to cutáneous vasodilatation alone this means an increase of 4·3 c.c. for 66 c.c. of skin or 6·5 c.c. per 100 c.c. skin. As will be seen shortly, body warming in the way described causes neither flushing nor warming of the forearm skin and it is clear therefore that the increase in blood flow must be relatively small ; it must be smaller than that giving rise to the flare. We have compared the increase of blood flow due to body warming with that due to the active hyperæmia caused by pricking in histamine or morphine at spots sufficiently numerous to provoke a bright flare over the whole forearm. This widespread flare raises skin temperature by 2 or 3°C[*] and blood flow
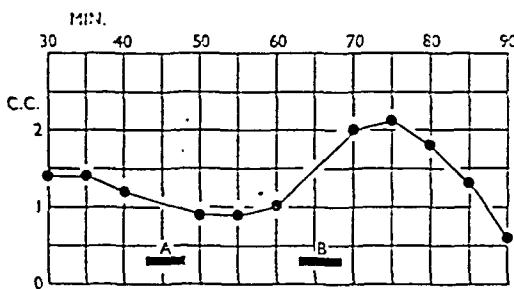
Fig. 6. Effect of flare on forearm blood flow. R.B.P. 11.11.36. Right forearm in cuff plethys-mograph ; forearm volume 620 c.c. ; bath temperature 30° ; room temperature 14°. Fore-arm blood flows measured with distal circulation arrested, and charted in c.c. per 100 c.c. limb volume per min. At A, between 43 and 48 mins., as a control measure, wrist cuff removed and plethysmograph slipped down to wrist, exposing upper forearm ; 10% morphine tartrate pricked into numerous spots in upper arm, producing a widespread flare ; sphygmomanometer and cuff replaced and blood flow readings resumed. At B, between 63 and 69 minutes same manipulations carried out except that morphine pricked into numerous spots all over forearm skin to be enclosed in plethysmograph.

to about 2 c.c. per 100 c.c. limb volume per minute, the average of 9 observa-tions on 3 subjects being 2.2 c.c.. These observations are illustrated in Fig. 6. In this example, taken from the same subject as Fig. 5, forearm blood flow increased from 6·0 to 13·2 c.c. for a forearm volume of 625 c.c. (11% skin). Calculation in terms of skin alone shows that the cutaneous blood flow was increased by 10·5 c.c. per 100 c.c. skin by the flare. This is considerably greater than that caused by body warming and is in keeping with the difference in skin colour and temperature. We have obtained similar results in a patient suffering from urticaria from cold ; the application of water at a temperature as high as 26°C provoked a full urticarial response

* According to Lewis and Grant (11), histamine pricked into the forearm skin raises skin temperature no more than 0·5 to 1·0°C, but we find the rise to be greater. The difference is due partly to the fact that we have provoked a wider spread flare by making numerous pricks over a large area of skin and partly to a difference in the initial skin temperature. In Lewis and Grant's (11) observations forearm skin temperature was already high (30 to 31°) when histamine was pricked in ; in our recent observations the forearm had been exposed for some time and skin temperature was lower (27 to 28°C).

from the forearm skin. In this patient as is shown in Fig. 7, body warming raised forearm blood flow from a level of between 0·5 and 1·0 c.c. to between 1·5 and 2·0 c.c., while as Fig. 8 shows, the temporary application of cold water to the forearm within the plethysmograph to produce a full urticarial reaction raised the blood flow to between 2·5 and 3·0 c.c. per 100 c.c. limb volume per minute. Here again the active vasodilatation in the skin due to the urticarial reaction causes a small increase in forearm blood flow which, however, is greater than that due to body warming.

In the fingers, where from X-ray examination we estimate skin and subcutaneous tissue to form about 50% of the volume, we have seen that body warming raises blood flow from the same low levels as in the forearm to as high as 20 to 30 c.c. per 100 c.c. finger volume per minute or in terms of
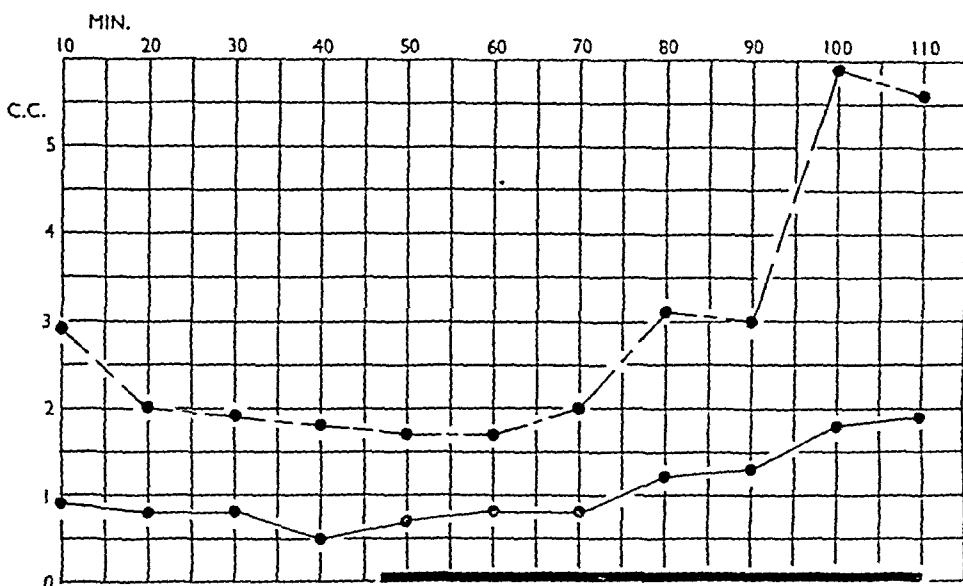
Fig. 7. Effect of body warming on forearm blood flow in a patient suffering from urticaria from cold. R.C. 24.11.36. Right forearm in cuff plethysmograph; forearm volume 600 c.c.; bath temperature 30°; room temperature 13°. After 47 minutes' rest body warming begun and continued throughout as shown by heavy line. Blood flows measured with distal circulation free (broken line) and arrested (continuous line) and charted in c.c. per 100 c.c. limb volume per min.. Compare lower curve with Fig. 8.

skin alone to 40 to 60 c.c. per 100 c.c. skin per minute. From these observations, even allowing for the small proportion of skin in the forearm, it is clear that the increase of blood flow provoked by body warming is much less than in the distal parts.

We have already mentioned that in contrast to the hand, the forearm skin is neither flushed nor warmed by warming the body. In this connection it is to be pointed out that just as venous blood returning from the hand exerts a considerable influence on the blood flow measured in the forearm when the distal circulation is free so it also modifies conspicuously the

temperature of the forearm skin ; to measure the surface temperature due to arterial blood supplied to the forearm alone, the circulation to the hand requires to be arrested. The earlier observations of Lewis and Love (13) on the effects of cold bear on this point. They showed that if congestion is caused and maintained in the arm, and the hand is immersed for a few minutes in ice cold water, the skin over those superficial veins which carry the blood away from the hand is cold ; this coldness spreads very slowly up the arm and the skin may be chilled in the line of the vein as far as the elbow. Further observation under more usual conditions, without congestion and the hand exposed to room air, shows that the venous return from the hand not only modifies but is a chief factor controlling the temperature of the forearm skin. The warming and slight flushing of forearm skin that occur
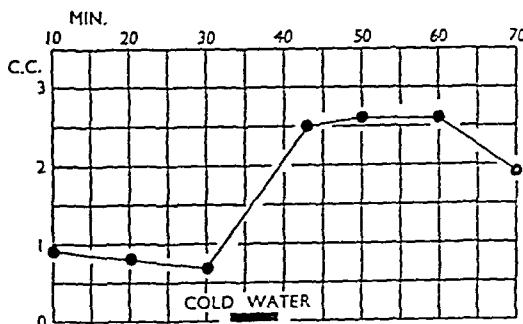
Fig. 8. Effect of urticarial reaction on forearm blood flow in same patient as in Fig. 7. Same conditions as in Fig. 7. Blood flow measured only with distal circulation arrested. After 33 minutes' rest, the warm water in the plethysmograph was drained off and water at 16°C was circulated for 5 minutes as shown by heavy line in the chart ; water at 30° was replaced at 38 minutes and blood flow readings resumed. The patient experienced severe itching of the forearm and at the end of the observation the skin within the plethysmograph was noted to be completely whealed. Blood flows charted in c.c. per 100 c.c. limb volume per minute.

when the body is warmed is due entirely to the venous return from the hand.* If the circulation to one hand is arrested, forearm skin on that side remains pale and cool as is illustrated in Fig. 9. Moreover, the warm venous blood returning from the hand comes mainly from the fingers. If the circulation to the fingers and thumb is arrested by constricting rubber bands, the skin temperature of the rest of the hand and of the forearm warms more slowly than when finger circulation is free and rises rapidly when the bands are removed.

These findings are consistent with the plethysmographic observations just described. They indicate that while body warming greatly increases the circulation in the hand and specially in the fingers, it causes no more than a slight increase in the forearm. Corresponding observations on the

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* It follows that the effect is greatest over the large veins and least most remote from them. Some areas may be found, depending on the distribution of the veins, in which venous return from the hand has no distinct effect on forearm skin temperature.

blocked. When 10 minutes' circulatory arrest is used, the flush and rise of skin temperature (no more and usually less than 1°C) which come quickly on both sensitive and anæsthetic areas on restoring the circulation are of the same degree and duration on both. We are unable to account for the slight differences in our results. It is clear, however, that the vasomotor effects of nerve block are strikingly greater in the distal than in the proximal parts of the limbs and correspond closely with the effects of body warming. We can therefore accept the conclusion that the differences are to be attributed to the distribution of the arteriovenous anastomoses.

Previous work (6) has shown that the anastomoses in the hands and feet are of chief importance in maintaining the warmth of these parts when exposed to cold ; but it was thought, in contrast to what was demonstrated in the rabbit (5) that, owing to their limited distribution, they could play but a small part in regulating body temperature. The new observations,
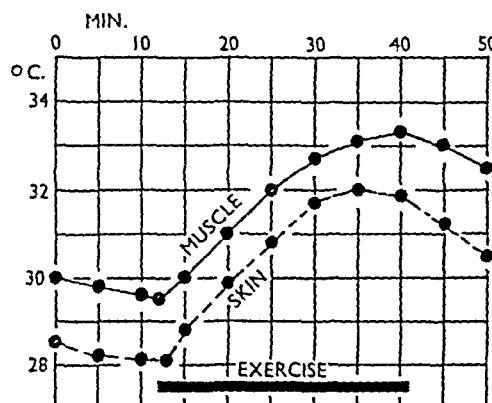
Fig. 10. Rise of skin and muscle temperatures due to exercise of forearm muscles. D.S. 12.10.36. Temperatures of skin over dorsum of forearm and from thermal junction in muscle 2 cm. beneath surface. Forearm cooled by exposure for an hour to room air at 15°C. Heavy line shows duration of exercise of lifting and dropping hand once a second.

however, extend the part played by these vessels in temperature regulation both local and general. It is clear that return of blood from the extremities through the capacious plexus of superficial veins of the limbs must exert a considerable influence in regulating body temperature ; the cooling effect of exposing and bathing the limbs in warm weather is a matter of common experience, and in the laboratory we know how readily body temperature is altered by immersing the limbs in hot or cold water. Again, the venous return must be effective in maintaining the warmth not only of the hands and feet but also of the more proximal parts of the limbs when the anastomoses are opened up by local cold ; it is to be remembered that the local vascular reaction to cold in the forearm and leg is insufficient by itself to maintain skin temperature.

Influence of muscle blood flow on skin temperature.

In connection with the foregoing observations we were led to enquire whether increased blood flow in the muscles influences materially the temperature of the overlying skin. It is well known that exercise provokes a great and long lasting increase of muscle blood flow. Eggleton (2) states that the skin over an active muscle may rise over 1°C, while Lewis and Pickering (14) record that exercise of the hypothenar muscles warms the ulnar side of the hand by as much as 2° to 3°C within ten minutes. It is not clear how this warming comes about.

We have already pointed out that in the forearm of the resting subject exposed to room air, skin and muscle temperatures fall progressively towards that of the room, muscle temperature being several degrees the higher and falling more slowly. If the muscles are exercised even gently, muscle temperature* soon begins to rise and is followed quickly and closely by a rise of skin temperature (see Fig. 10) though there is no appreciable flushing of the skin. The rise is steeper the more vigorous the exercise;
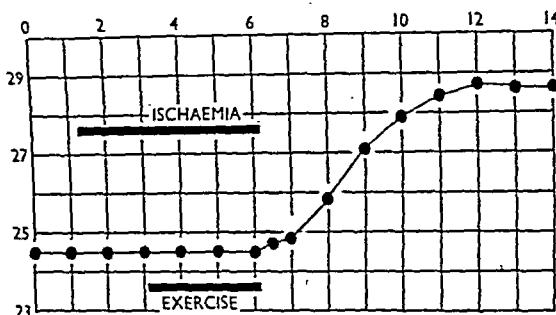
Fig. 11. Rise of skin temperature following exercise and ischæmia. R.T.G., 13.10.36. Skin temperature from dorsum of right forearm over belly of extensor muscles. Forearm cooled by exposure for an hour to room air at 15°C. Upper heavy line indicates duration of circulatory arrest at upper arm. Lower heavy line shows duration of exercise of lifting hand every second. At 10 minutes the flush of the reactive hyperæmia was much faded.

skin temperature may rise as much as 5° in as many minutes. In some of these observations we have taken the precaution of arresting the circulation at the wrist to prevent any possible effect of blood returning from the hand on forearm temperature. This is unnecessary; the hand already cold does not become warm and arresting its circulation does not alter forearm temperature. By testing with the finger or a movable thermal junction it is found that the increased warmth of the skin with local exercise is confined to the neighbourhood of the active

* For these observations we have found it better to use not a needle thermal junction but one of fine (32 gauge) enamelled copper and constantan wires soldered together at their tips, such as we use for recording skin temperature. A wide hypodermic needle is inserted into the flexor or extensor muscles and through this the wires are passed. The needle is withdrawn over the wires, leaving them within the muscles. This flexible type of junction causes less pain than a rigid needle when the muscles are exercised and when introduced parallel to the muscle fibres usually gives rise to but slight discomfort.

muscles and decreases rapidly beyond their boundaries. For example, when the anterior tibial muscles are exercised by repeatedly lifting the foot, the temperature of the overlying skin rises rapidly and to a high level; over the tibia the rise is but slight and delayed,* and is absent in the calf. The increased warmth of the skin is not due mainly to a concomitant cutaneous vasodilatation but to heat conducted from the underlying muscles. The skin does not flush but rather pales. If an area of skin is blanched by adrenaline introduced intradermally, the rise of temperature provoked by exercise comes as usual; it is probable, however, that a rise of skin temperature of this order increases skin circulation to some extent by the direct effect of warmth on the skin. Further, the rise of temperature is not due mainly to increased heat production within, but to increased blood flow through the working muscles. If the muscles are exercised while the circulation is arrested, skin temperature does not rise until the circulation is restored and then rises although exercise has already ceased. The exercise of lifting the hand, for example, can be maintained for 3 to 4 minutes before the ache that develops becomes intolerable; as is shown in Fig. 11, the skin temperature begins to rise steeply within 10 seconds of restoring the circulation at the end of exercise. The reactive hyperæmia following ischæmia for this period gives by itself no rise of skin temperature. The conspicuous rise of temperature following exercise and ischæmia long outlasts the skin flush.

We will not pursue here these observations on the vascular effects of muscular activity but we wish to point out that they show clearly that direct conduction of heat from working muscles is an important factor in warming the overlying skin. It would seem that this factor must play a greater part in the upper portion of the forearm and leg, where muscle predominates, than in the lower which is more bony and tendinous; it is absent from the digits. Less directly, however, muscular activity does influence the temperature of the digits as Lewis and Pickering (14) have shown. They point out that the raised temperature of active muscles must warm, and thus tend to dilate neighbouring arteries passing to more distal parts.

Summary.

1. Observations are recorded on the human arm and leg showing that differences exist between the circulation in the extremities and that in the more proximal parts.

2. In measuring change of skin temperature and blood flow of the proximal parts precautions are required to exclude the effect of venous blood returning up the limb from the extremities.

3. While in the extremities sensory stimuli applied to the body cause vasoconstriction they have either no effect or cause vasodilatation in the forearm and leg.

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* The inference is that exercise provokes but little, if any, change in bone blood flow.

4. Small doses of adrenaline regularly cause increase of limb volume and blood flow in the forearm and leg, due to vasodilatation in the voluntary muscles. The vasodilator reaction to adrenaline is increased after sympathectomy.

5. In man, as in other animals, circulating adrenaline is not the factor responsible for the regain of vascular tone following sympathectomy.

6. The vascular changes in the distal and proximal parts of the limb are described in response to alterations of body temperature.

7. Increased blood flow through active voluntary muscle is shown to exert a conspicuous effect (by the conduction of heat) on the temperature of the overlying skin.

8. The effects of body warming and of muscular activity are discussed in relation to their bearing on the maintenance of body temperature, both local and general.

9. The arteriovenous anastomoses of the extremities play a greater part than was hitherto realised in maintaining the temperature of the limbs and in regulating body temperature.

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EFFECTS OF ASPHYXIA AND PRESSURE ON SENSORY NERVES

OF MAN.

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WHILE investigating the effects of asphyxia upon pain responses in the human skin (Lewis and Pochin (7)) it became clear to us that, although a number of observations had been recorded previously (1, 4, 5, 6), our knowledge of loss of various forms of sensation in the skin and underlying tissues during asphyxia still remained incomplete. The observations here recorded were undertaken in the first place with the idea of filling in these gaps ; it seemed important that this should be done if only to assist the many recent attempts to correlate different sensory functions with different physiological types of nerve fibre. The observations originally planned on asphyxia are recorded in Part I ; some observations upon an unexpected sensory paralysis in a finger are added in Part II.

PART I. EFFECTS OF ASPHYXIA ON SENSORY NERVES.

Our tests of cutaneous sensibility have been made for the most part upon the fingers, and almost always upon the dorsal surface of the terminal phalanx, between the nail and the interphalangeal joint. During the period of asphyxia the hand has been kept in water at 35° and the tests have been conducted upon skin at that temperature, except in the case of the response to warmth when the temperature of the tested finger has been kept between tests at 30°. Before skin which is to be tested finely and repeatedly is immersed, it is thinly smeared with vaseline to prevent the skin imbibing water.

The apparatus used for testing touch and pain has consisted of Frey's hairs of varying and suitable thickness, and needles mounted on similar hairs. In testing cold and heat spots a mass of copper ending in a tiny cylinder has been used ; for massive thermal stimuli larger metal surfaces have been found convenient ; in each instance the metal is withdrawn from a water bath of desired temperature, dried and at once applied to the skin.

Another form of massive thermal contact has been a small beaker of water brought upwards to immerse the dependent finger tip. To measure the delay in the response to thermal stimuli, the metal is brought against skin over which a fine wire is stretched, the instant of contact being recorded electrically ; where immersion in water has been used, immersion has been abrupt and has been signalled electrically.

In asphyxiating we have used one of three methods. Usually we have employed the pneumatic cuff, encircling the upper arm. This is the most satisfactory plan when relatively short periods of asphyxia are desired, namely up to 30 min., as when the early changes in sensation are investigated. But this method asphyxiates not only the nerves under the cuff, but all the tissues of the limb including nerve endings ; and for this reason we have repeated all the chief observations, using the special clamp described in a previous article ; this clamp only renders those parts of the nerves ischæmic which lie within it, the circulation to the arm below the clamp remains free. Thus effects of distal ischæmia are excluded.

Because it is necessary, in attempting to abolish all sensation in the fingers, to continue asphyxia for very long periods, and because it is unwise repeatedly to render the whole limb or its main nerves ischæmic for such periods, in investigating the last stages of sensory loss we have asphyxiated the finger alone. To do this a band of rubber, 2½ cm. wide, is wrapped firmly in several overlapping turns around the base of the finger. If a narrow band is used, deforming pressure is introduced and the effects cannot be regarded as simply asphyxial (see Part II). When the broad band is used sensation in the skin of the finger is completely lost in about 2 hours.

Touch. The first obvious sensory defect noticed when the circulation to a limb is stopped is in the sense of touch ; it consists of numbness of the very tips of the fingers. It has been shown in previous work (6) that this occurs in about 13 to 15 min. when the obstruction is at the upper arm and the arm is kept at 35°., but that it comes at later times if the obstruction is placed lower on the limb. If the obstruction is around the base of the finger it comes at about 25 to 30 min.. This numbness seems to be not only the first obvious change, but actually the first sensory change in the skin consistently to be detected. If the threshold of the skin to touch is tested with Frey's hairs of suitable strength, numbness can be detected as early as, and sometimes a little earlier than, the first sign of raised threshold to hairs. Vibration sense, tested with a 512 tuning fork at the base of the nails, is impaired several minutes before, and is lost when, numbness appears. But this is only the case when the circulation to the fingers has been arrested ; when the special clamp is used vibration sense begins to decline with the onset of numbness or a little later. Because it seems clear that a local factor interferes when the circulation to the skin is arrested, we are unable to regard the early impairment of vibration sense as significant, so far as loss of function in the nerves is concerned. To resume, from the time when numbness comes, the threshold to light touch rises and numbness steadily increases in degree

until all sense of touch is lost. By the time sense of superficial touch is lost, sense of deep pressure has also gone. If the upper arm has been compressed, the skin of the finger tips becomes anæsthetic in about 20 to 25 min. ; if circulation has been stopped at the base of the finger, anæsthesia is complete at the tip after about 35 to 50 min..

It will be most convenient to express the onset of defects in the other forms of sensation by relating it to the onset of the first defect in touch. Loss of the other forms of sensation will be related to anæsthesia and supplemented by statements of times.

Cold. The time at which a sense of coldness to cold contacts is noticed to change depends upon the method used to elicit it. The earliest defect in the cold sense may be detected by mapping out a number of cold spots on the dorsal surface of the terminal phalanx of a finger and testing these (while at 35°) at regular intervals by short contacts with a small metal surface at 10°. The responses begin to fail at the same time as, or a minute or two before or after, numbness is first noticed in the tested skin.

If the tip of a finger, which has been lying in water at 35°, is immersed in water at 12°, or its dorsal surface is tested with a larger metal contact at similar temperature, the first defect in the cold sense during asphyxia is noticed as a delay in the first appreciation of cold or as a delay in the full development of the sensory response. The reaction time often begins to widen when numbness has first developed in the tested skin, or the delay may not be detected till anæsthesia appears ; the sensation of cold when it comes after a delay, increases with unusual gradualness to its height. If the first clear sense of coldness is signalled, then the original reaction time of 0·4 sec. on the finger widens out ultimately to as much as 5 or 10 sec.. This increased latency is observed with asphyxia of the whole arm and equally well when asphyxia is confined by means of the special clamp to the nerves in the upper arm. If attention is directed merely to the degree of coldness. to the exclusion of the time factor, then the cold sense will not be recognised as affected until about the time when anæsthesia develops, for until then the ultimate intensity of the response is usually undiminished. Response to cold though progressively delayed, and often diminished, is not abolished till long after anæsthesia is full. In observations upon the asphyxiated finger it is lost at about the 110th minute and, as we shall see, before pain.

In testing the finger by immersing it, we have found that the asphyxiated finger develops a painful response to cold, which will be discussed later. Coming, as it does in the final stages of asphyxia, pain occurs as soon as 3 to 6 seconds after the finger has been immersed in water at 15°, and in the late stages of asphyxia it may interfere with tests for cold response. appearing before or replacing the latter.

In considering the delay of the cold response, this is not comparable to the delay of the pain response when the first response fails (see next page). The interval is much greater in its duration. The second pain response on

the finger is delayed by at most 1·5 sec. ; the cold response may be delayed by 10 sec.. Further, the latency for cold perception increases gradually from 0·4 to many seconds, while for pain the latency rises abruptly from 0·3 to 1·5 sec. where it remains constant. Clearly, the cold response, unlike the pain response, is not carried by two groups of nerve fibres of widely different conduction rates. Testing the warm foot with cold metal, it has not been possible, however short the contact, to display two responses. The quick decline of the response of the normal skin to a short cold stimulus, and the long delays which occur in the response to cold in the later phases of asphyxia would suggest that these delays are not due to the existence of slowly conducting cold fibres, for the rate involved would be so extraordinarily slow. Similarly it seems unlikely that so great a delay is due to slow conduction in the stretch of fibres asphyxiated. The delay may be due to a greater time needed either for cooling deeper nerve endings, or for fuller cooling of the superficial ones, to provide impulses adequate to pass the asphyxiated nerve fibres.

Warmth. As with cold, the times given for loss of warm response will vary with the method used. If warm spots are mapped out on a finger maintained at 30°, and these spots are tested by short contacts with a small metal surface at 40°, the responses are found to fail first a little after the tested area becomes numb. If a massive stimulus is used, the finger tip being transferred from water at 30° to water at 42° or 43°, the first defect noticed is delay in the warm response ; exceptionally this may be recorded as early as the onset of numbness ; usually it is recorded at about the time anæsthesia develops. This delay, mounting as it may from about 0·8 sec. to 5 or 10 sec., is also found when the special clamp is used, and presumably arises similarly to the delay in response to cold.

The warm response is noticeably diminished from the onset of anæsthesia, it is lost at the same time as, or a very few minutes after, the cold response, responses to pain being still detectable.

Pain. This has been tested with needle points fixed upon hairs of different bending strain, and in a previous article we described the twofold response, which occurs to a single needle prick ; in the present observations we have paid attention to both first and second (or delayed) pain response described by us in the recent paper (7). The recognition of the first and second pain response to needle prick comes most easily—and familiarity with it is necessary before observations can begin—if the fine needle is mounted on a hair of about 0·2 g. bending strain. If this is applied for an instant to the thin skin on the front of the wrist, usually the first or immediate response will fail and the second response is clearly perceived after the needle point has left the skin. Incidentally, in the method which Frey (2) used in testing, namely, pressing the needle point on the skin until pain is felt, it is usually the second and not the first response which appears. The second is elicited by lighter stimulation than the first response, and is longer lasting. Sometimes, and especially when the response is excessive,

it may last for as many as 10 or 15 sec. ; it then gives rise to a sense of local irritation, which is relieved by lightly rubbing the skin. This aftermath or prolonged sting is especially well felt on the skin of the face, but here, owing to fusion of first and second responses, it begins at once after the immediate prick of the needle. The prolongation of sting has been noticed by previous workers and in calling attention to it again we emphasise that it belongs not to the immediate, but to our second pain response.*

If pricks are used during asphyxia such as are just adequate usually to provoke an immediate response from the back of the finger, then we find that this first response is substantially unchanged after numbness has developed, it begins to fail after the skin has become conspicuously numb ; and it is lost well after the skin becomes anæsthetic. The same events are noted whether the cuff or the special clamp is used on the upper arm, or the finger is tied off. But if the strength of the stimulus is much increased, the immediate response to the jabs of the needle is not lost until much later. These responses are finally lost at about the times when the cold or the warm response vanishes.

With light pricks the second response becomes exaggerated, being more intense and conspicuously long lasting, a little before the onset of anæsthesia. The second responses do not diminish in number or intensity until long after anæsthesia has become established (or about an hour after the finger has been asphyxiated from its base) ; and the delay in the response remains unchanged. The responses are not all lost until after all cold and warm responses have disappeared, though by this time the second responses are diminished both in number and intensity. They are usually all lost within 10 or 15 minutes of the loss of all responses to cold and warmth, or at about 120 to 130 min. in the asphyxiated finger.

Pain sense in deeper structures is still present very late in asphyxia. It may be tested by squeezing the web, which lies between two fingers, or by gripping the muscles of the thenar eminence and exerting pressure on them. Each of these procedures gives rise to the corresponding and characteristic pain.

Sense of position and muscle pain. As stated in a previous paper (6) sense of position in finger or hand is lost when anæsthesia reaches the corresponding joint level. This relation requires no further comment at the moment. We proceed to consider a special point raised by Matthews. He has suggested (8) that the pain, which has been shown in this laboratory to be derived from muscles when these are worked under ischæmic conditions, may be conveyed by afferent fibres connecting to muscle spindles. We have thought this to be improbable, and have been inclined to think of afferent pain impulses on the one hand, and of afferent impulses regarded as conveying information relating to muscle tone on the other, as almost certainly requiring specific and distinct channels of conduction. The

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* It may here be recorded that the aftermath or sting is not produced by circulatory arrest and is not attributable therefore to the release of a stable pain-giving substance in the skin .

matter has been tested further in conjunction with tests of sense of position in the asphyxial experiment. The pain provoked by squeezing muscle and that provoked by working it under ischæmic conditions are in fact indistinguishable ; but the latter cannot be provoked so late in asphyxia as the former, because not long after palsy occurs in the motor fibres of the nerve trunks (approximately simultaneous with complete loss of function of the touch nerves), the response of the muscles to a faradic current directly applied becomes weaker and eventually it ceases. It is quite easy to show that pain provoked by squeezing the thenar muscles is present long after all sense of position in thumb and even in wrist is lost. This comparison however is insufficiently searching, because the loss of position sense applies only to passive changes and occurs when the muscles have reached a flaccid condition. If the muscles of the thenar eminence are faradised at the period of asphyxia when the hand is almost completely anæsthetic to the line of the wrist, these muscles can still be made to contract quite strongly ; and, if the time and the point of stimulation are well chosen, little or nothing will be felt except a clear sense of tension in the region of the muscle contracting. This sense of tension will often give an idea of the movement which has happened in the thumb as long as 3 or 4 minutes after a passive change of position has become unrecognisable. A little later the sense of tension on faradising the muscle is itself lost and there follows a period during which the muscles can still be made to contract, at first fairly strongly, but as time passes less strongly ; they then contract without the subject being in any way conscious of the fact. When these contractions have been continued for a minute or two (the duration of stimulation required to produce pain at this time may be shortened if the same muscles are submitted to 1 minute of preliminary stimulation at an earlier stage of asphyxia) pain develops and continues ; it is felt in the region of the ball of the thumb and is characteristic in its quality ; the muscles become tender in that the same pain is now very easily exaggerated by simple pressure and continues to be for a period during which it may be thoroughly tested. These observations make it clear that pain, coming from muscles working under ischæmic conditions, or exaggerated by light pressure upon muscles that have been worked, can occur not only after all sense of position in the relevant parts is lost, but also after all those afferent impulses are lost, which are responsible for the subjective local sense of tension to which contraction of muscle gives rise. Thus, our view that the muscle spindles are unconcerned in the production of pain derived from muscle is strengthened.

It is here to be remarked that the asphyxial experiment distinguishes two factors affecting recognition of position. Firstly, there is a factor, which very possibly is touch and deep pressure sense, and which is probably the dominant one when the hand is normal. Secondly, there is a sense of tension, which develops in the region of the muscles concerned and aids recognition of changed position when muscular contraction is responsible for it ; this disappears later in asphyxia than the first factor.

Hyperalgesia. We have given special attention to the development of hyperalgesia. It develops when the whole arm is rendered ischæmic, when the finger is rendered ischæmic, or when the nerves of the upper arm are compressed by the special clamp while the circulation to the hand remains free. In all instances it comes simultaneously with, or within a minute or two of, the appearance of distinct numbness and it is readily displayed by light friction, or by squeezing the finger lightly, or by depressing the nail ; the skin is sore to friction, and squeezing the tip of the finger or pressing on the nail gives much the same painful response as is obtained from a bruised finger. This hyperalgesia is one which concerns both the first and the second response. Thus a single short rub of the skin is accompanied by distinct though slight pain, and depression of the nail is immediately painful. These immediate painful responses are the first to be noticed, but they decline as the skin of the finger becomes anæsthetic and a delayed response then becomes manifest. At this stage a short rub of the skin, or a short pinch of the nail, is not painful at the moment of the stimulus, but is followed about a second after by a short and very obvious pain. This response on the finger and on the toe has the same delay as has the second response to needle prick of the same part. Simultaneously the first response to needle prick is declining while the second response is manifestly exaggerated, the pain being more intense, more diffuse, and longer lasting. We emphasise that, although the second pain may be unpleasant, it is never associated with any distinct reference ; it is localised.

The second response becomes exaggerated from the onset of anæsthesia and remains exaggerated until the later stages of asphyxia, when the decline of pain sense occurs rather rapidly. Curiously, however, the second response to pinching the nail may remain very painful when the skin is no longer responding at all to needle prick. This delayed extinction of function is presumably in deep lying pain nerves.

During the stage of asphyxia in which numbness is appearing in the finger tip, the response to 15° water begins to acquire a very distinct sting after about 30 sec. immersion. From this time onwards immersion at 15° is found to yield an obvious stinging pain at shorter and shorter intervals, until at the end of the asphyxial experiment, the delay may be as short as 3 sec.. On several occasions this stinging response to cold water has been noted at a time when the skin was quite insensitive to needle prick. Very possibly it also is a response by deep lying pain nerves. When the tip of a warm finger is similarly immersed immediately after arresting the circulation to it, the stinging pain just described does not develop, even if the finger is kept immersed for a minute or more.

A similar pain reaction is observed if warm water (39° to 41°) is substituted for the cold during the long asphyxia.

The order of sensory cutaneous loss.

If we consider the order in which the various sensory functions of the skin are lost in asphyxia, enquiry among past records (Herzen (5), Fabritius and Bermann (1), Goldscheider (4), Lewis, Pickering and Rothschild (6)) find these to be in general though incomplete harmony. There is agreement that touch is lost first, and that cold sense, warm sense, and pain follow. But the precise order in which the last three are lost has remained uncertain. Actually the form of statement is inadequate. We require to know not only when these functions go, but the manner of their going. Our present studies have explored this aspect particularly and our results may be summarised.

The first obvious defect is in touch sense ; numbness appears and this defect is as early as any we can consistently detect. Simultaneously or within a very short time both cold and warm sense begins to be affected. The defect in all three is progressive from the start. The defect in touch progresses most rapidly and this sense is the first to be lost. About the time
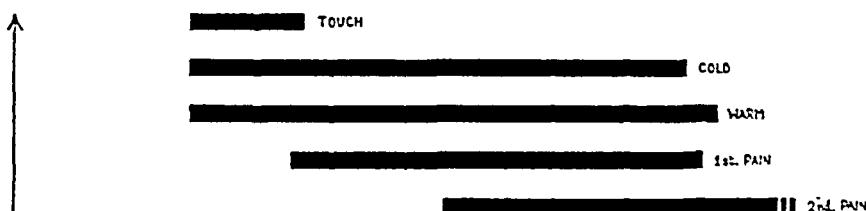
A diagram, to be read from left to right, indicating the approximate time relations of loss of various forms of sensation from skin during a period of asphyxia, which begins at the arrow.

when it is lost, the first response to pain begins to diminish and at a later period a similar defect appears in the second pain response. The defects in pain sense are preceded at first by exaggerated response ; their decline once it starts is progressive. Decrease in appreciation of cold, warmth, and pain are all very gradual and continue long after touch sense has completely gone ; the first stages of their decline are so long drawn out that the precise times of their individual loss is not always easy or possible to establish. But it may be said that loss of cold sense, first response to pain, and warm sense, occur almost simultaneously. Probably the usual order of loss is that stated, though this cannot be said to be invariable. Loss of the second response to pain comes later than the others, but may not be delayed long in skin ; complete loss of pain sense to needle prick has, however, not often been recorded, and where it has, a second pain response to pressure on the nail remains.

The decline in the sensory functions considered may be expressed in order to facilitate understanding of the broad time relations, by means of the accompanying diagram. The essential points are that defects in touch,

cold sense, warm sense, and first pain response begin almost simultaneously. Diminution in second response to pain is alone much delayed. The only sense to be lost early is touch, and from the times when this occurs the decline *in all remaining senses is steady to elimination* ; the times of elimination of all these fall within a relatively short time period. Thus, it would be mistaken *to think* that the different senses decline over distinct successive periods, they all decline together, but touch vanishes first, and pain is the most resistant. It is also to be emphasised that the periods of decline of first and second response to pain, the first conveyed by fast and the latter by slow conducting fibres, do not occupy separate phases of asphyxia, they overlap considerably and the end of these two periods are not widely separated in time.

Relation of function, rate of fibre conduction, and fibre size.

The original idea that nerve fibres are put out of action by different agencies according to the rates at which they are capable of conducting, is already acknowledged to form but an imperfect correlation (Gasser (3)) ; there is no clearer example of this than the influence of asphyxia upon fibres conducting pain impulses. There are, as we have shown previously, two chief groups of pain fibres, conducting at rates of very different order ; but under asphyxia, elimination of function in these two sets of nerve fibres is very gradual, and in large measure simultaneous. It is true that the faster conducting pain fibres begin to drop out first, but these are not all eliminated, until the elimination of function of the slow conducting fibres is also far advanced.

An attempt to determine with useful degree of precision a relation between different sensory functions and size of nerve fibre, seems also doomed to failure, if we are to accept the effects of asphyxia as a basis for such correlation. The more closely the examination is conducted, the more obvious is overlapping. It is true that touch is most heavily affected in the early, and the second pain in the later, stage of the asphyxial experiment ; but touch begins to be affected no earlier than sense of cold or warmth, and the period of pain elimination does not lag far enough behind to escape wide overlap with the periods corresponding to the elimination of each of the remaining sensory functions. We are not surprised to find Gasser concluding in his recent publication that fibres subserving deficient sensory functions must be widely distributed through various fibre sizes ; our recent experience tends indeed to emphasise rather than to diminish the difficulties confronting attempts to correlate in any simple fashion sensory function and fibre size.

PART II. PRESSURE BLOCK OF DIGITAL NERVE.

When a 6 mm. wide rubber band is wound tightly around, and fixed in place upon, the base of a finger, the manner in which sensation is lost in the finger beyond the band is similar to that described for a finger asphyxiated by the use of the broader band. The order in which diff. t kinds of

sensation are lost is the same, and the times are the same ; thus all sensation is lost or practically lost in the finger kept at 35°C. at the end of 2 hours.* But when the narrow band is removed, it is found to have cut deeply into the tissues of the finger and, although bloodflow returns promptly, sensation may not.   On two occasions sensation has failed to return for a period of weeks or months to one side of the finger, indicating that a pressure paralysis has happened in one of the two digital nerves.

This lasting condition of sensory loss in a finger has occurred once in each of us.†   The loss of sensation was partial, sense of touch alone being depressed ; but pain sense in each instance was noted to be exaggerated.

In the case of the first persistently numb finger (E. E. P.) observed, it was noted that the numbness tended to disappear in the morning, and to return in the evening.   In the case of the second numb finger (T. L.), occurring after the first had fully recovered, a similar but quite independent observation of diurnal variation was made.   The dual observation stimulated us to investigate the second instance more closely.   The observations on this finger follow.

Hypoœsthesia.   The finger was affected along its ulnar side from the proximal interphalangeal joint to the finger tip ; it was very numb, but never quite anæsthetic in any part.   When morning improvement was first noticed it was thought to represent the beginning of recovery ; but by evening all that had been gained was lost.   These changes were repeated on subsequent days ; it was noticed too that in the morning the finger was colder than in the evening.   This led to deliberate tests of the finger, after immersing the hand in warm (39°) or cold (20°) water.   In numerous tests in which the skin was tested by means of Frey's hairs of different weights it was shown that immersion at these temperatures for 3 minutes invariably changed the capacity of the affected finger to recognise touches. Using similar temperatures we could detect little or no change in the tactile perception of adjoining fingers.   On the affected finger, when warm, touches with light hairs usually failed to be appreciated and with heavier hairs were felt faintly, while on the affected finger, when cool, touches with light hairs were usually felt and with heavier hairs were felt plainly.   In a large number of further observations different parts of the affected finger were tested, cold water being circulated through a capsule in each case for 3 min. around the tip of the finger, its middle section, or its base, while the hand as a whole was immersed in warm water ;   or in other instances warm water was circulated locally while the hand as a whole was immersed in cold water. These tests, some of which were done with the circulation to the limb free and others with the circulation arrested, gave consistent results ;   arrest of circulation was introduced to ensure that change in temperature of one part

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* We refrain from concluding that the order of paralysis is in this instance governed by pressure, because clearly asphyxia is also concerned.

† And caused us to abandon the method of rendering the finger bloodless with a *narrow* band, owing to the long-lasting inconvenience resulting.

of the finger was not conveyed to another part. We were unable to demonstrate that change in the temperature of the skin over the terminal phalanx, where the testing of touch was always done, or of skin of the middle part of the finger, had any effect on the recognition of touches. On the other hand cooling and warming the base of the finger, the region to which the pressure band was originally applied, always produced definite effects, and similar to those produced by cooling and warming the whole finger or the whole hand.

These tests made it abundantly clear that the improved touch sense observed in the cold finger depended upon the temperature change at the region of injury. The improvement was seemingly the result of cooling in the region where the nerve originally suffered pressure and damage, and was due neither to cooling of the skin receiving the touch stimuli, nor to any change in the temperature of the nerve conducting the impulses from tip of finger to the region of injury. It sufficed to place a flattened lead tube (6 mm. in width) against the ulnar side of the affected finger at its base and to allow a current of water at 20° to flow through this, to produce a definite improvement in the tactile perception of the affected finger, while the whole hand lay immersed in water at 39°. Similar cooling of the radial side of the same finger was without effect. The effects of temperature change of the skin was tested over the range 13° to 42°. Little change in sensation was detected up to 27°, but from about this point to 42° the effects were definite and progressive.

Hyperalgesia. In the first instance of nerve injury from pressure, the numb area was first noticed to be displaying hyperalgesia the same evening. The change noted was a conspicuous and persistent second response to needle prick ; the first response was judged to be impaired. Pain was not again tested. In the second instance, hyperalgesia though repeatedly *noticed* previously, was first recorded as present about 3 weeks after the injury. This hyperalgesia was easily elicited by light friction and in area corresponded closely with that of numbness. The ulnar side of the nail and pulp of the finger gave obvious tenderness on pressure. To needle prick both the first and second pain responses of the skin were exaggerated, but especially the latter, which was also prolonged, sometimes for 10 or 15 seconds. Tested with copper at 40° to 42° no definite change in the threshold of the pain nerves to heat could be determined.

It was noticed that the hyperalgesia varied with the degree of numbness in the finger, being most conspicuous when the finger was most numb, and inconspicuous or absent when numbness was slight. The effect of temperature of the finger on hyperalgesia was therefore investigated in the same way as had been done for numbness. The hyperalgesia was found to be clearest when the hand was warmed, and to be almost absent when the hand was cold ; these effects were obtained equally when the heating or cooling was confined to the proximal phalanx. Thus when the whole hand was warmed in water at 39° for 3 minutes and then tested, it was found that pain was

regarded exaggerated pain responses as coming from pain nerves of a special order, and in others as coming from pain nerves in a special state, namely, influenced by degeneration or by regeneration.

The simplest method of which we know, which suffices to dissociate touch and pain is that described in the first part of this paper ; in asphyxia touch becomes defective before pain sense. It is very remarkable that in the phase of asphyxia in which touch becomes defective, hyperalgesia appears. The coincidence is striking and uniform. This again might be regarded as a disclosure of pain by withdrawal of touch ; or the hyperalgesia might be interpreted as due to a preliminary irritation of the pain nerves before function in these declines. There is no evidence which will conclusively fortify or destroy either of these views, and discussion could only proceed upon a hypothetical basis. Enough has been said perhaps to emphasise the striking but as yet unexplained association between hypoæsthesia and hyperalgesia, and to draw attention especially to this association in asphyxia. For these manifestations of asphyxia are very easy to produce in the human limb, are easily reversible and capable of repetition, thus lending themselves to further exploration.

SUMMARY AND CONCLUSIONS.

1. In asphyxia of a limb defects of touch, cold sense, warm sense, and fast conducted pain begin almost simultaneously. The defect in slow conducted pain comes later and is preceded by exaggerated pain response. The only sense to be lost early is touch; the rest all decline slowly and together and are eliminated within a relatively short time period of each other.

2. The declines of fast and slow conducted pain do not occupy separate phases of asphyxia, they overlap considerably. In asphyxia of a limb, the separate sensory functions are not eliminated in a simple precise order, neither do nerve fibres fall out in the order of their conduction rates.

3. Recognition of position resulting from passive movement, fails early and at the same time as touch and deep pressure sense fail. But sense of tension associated with muscular contraction remains until later and continues to inform the sensorium of changes of position brought about by such contraction. Thus there are two factors affecting recognition of position.

4. Pain sense in deep seated structures such as muscle is preserved until the very late stages of asphyxia and until after all sense of position is lost. Nerve fibres conveying pain impulses from muscles, and those conveying sense of position or of local tension, are separate.

5. An instance of pressure injury in a digital nerve is described. The injury gave rise to hypoæsthesia and hyperalgesia, which were found

regularly to be exaggerated by warming and diminished by cooling the region of injury. The meaning of these phenomena is discussed briefly, as is the curious association of hypoæsthesia and hyperalgesia in this and other circumstances.

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OBSERVATIONS ON THE BLOOD CIRCULATION IN VOLUNTARY MUSCLE IN MAN.*

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In a previous paper Pearson (11) and I described a plethysmographic method suitable for studying blood flow in the voluntary muscle of the human limb. We there dealt only briefly with some of the circulatory phenomena of active muscle, and paid chief attention to differences in the vascular responses to various stimuli displayed by the proximal and distal parts of the limbs. I now record observations† on the vascular changes accompanying muscular activity. These are of interest since our present knowledge is based almost entirely on animal experiment involving the use of anæsthetics, more or less extensive dissections and usually section and electrical stimulation of the motor nerves.‡ A number of plethysmographic observations has been made on the human arm (for example by Mosso (15), Athanasiu and Carvallo (3), Hewlett and Zwaluwenburg (12), Lewis and Grant (14)), but these do not exclude concomitant changes in the hand which may differ in extent and direction from those in the more muscular part of the limb. The use of the forearm plethysmograph with arrest of the distal circulation overcomes this difficulty. The new observations are of interest also for the investigation of peripheral vascular disease. This has been much studied in recent years but chief attention has been paid to the hand and there is yet no adequate method for assessing the circulation in the more proximal parts of the limbs and specially in the muscles. Though the observations that follow deal mainly with the normal, it will be clear that they form a basis for the further study of the abnormal.

It has been shown (11) that the proportions of skin and bone in a lean and muscular forearm are small (muscle forming about 85% of its volume), and that a cutaneous hyperæmia even so active as that of the flare causes only a relatively small increase of forearm blood flow (from 1·0 to 2·2 c.c. per 100 c.c. limb volume per minute) ; this hyperæmia is apparently greater than any occurring in the skin as a result of exercise of the forearm muscles.

Also, observations have been recorded to show that exercise provokes but little, if any, change in bone blood flow. It seems, therefore, that any material departure from the resting forearm blood flow resulting from local exercise can safely be attributed to a change in the muscle circulation ; this conclusion is supported by other observations. It is to be remembered that blood flow measured by Hewlett and Zwaluwenburg's method (12) must include also lymph flow, which is known to be increased by exercise. From the data available, however, (Barcroft and Kato (4), Drinker and Field (6)) lymph flow appears to be so much less than blood flow that it may be neglected without material error.

The details of method already published are sufficient for the arm at rest ; a few additional are required for exercise. The exercise used throughout is a strong grasping movement of the hand. This throws into contraction the large majority of the forearm muscles leaving only the relatively small pronators and supinators not fully employed, so that blood flow stated in terms of limb volume must nearly represent that through active muscle alone.* Since limb volume changes are to be interpreted in terms of vascular changes it is necessary to take into account and to reduce as far as possible those due to muscular contraction and relaxation. They are reduced to a minimum when the arm is arranged as follows. The elbow and wrist are supported on sandbags which must not touch the plethysmograph. The wrist cuff is so applied that when inflated its upper end is separated by a narrow gap from the lower end of the plethysmograph. The back of the hand is up and the fingers and thumb are loosely arranged round a horizontal wooden bar fixed to the water bath, so that when the bar is firmly grasped there is little or no displacement of the arm. To record contractions a small cuff is wrapped round the bar, connected to a recording tambour and inflated to a pressure of about 100 mm. Hg. The grasping movement can be maintained or made repeatedly without materially disturbing the rest of the body. The amount of exercise that can be undertaken with the wrist cuff inflated is limited by the ache that develops in the hand and lower forearm. When longer exercise is required the cuff is not inflated until towards the end of the exercise. A tolerable ache in the limb under observation or in the other limb causes no material change in blood flow.

To observe the changes due to muscular movement alone, the arm is first depleted of blood by applying a rubber bandage tightly from the fingers to above the elbow and inflating a wide sphygmomanometer cuff on the upper arm to well above systolic pressure. The bandage is removed and the arm arranged as just described. When the bar is quickly grasped and released the volume recorder oscillates momentarily ; these oscillations are largely avoided by making the movements more slowly (Fig. 1). Grasping

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* Accepting that muscle forms 85% of limb volume and assuming that 90% of the muscle fibres are contracted, blood flow rates stated in terms of limb volume require to be increased by one-third to represent more nearly that through muscle alone, (bone and skin blood flow being neglected).

the bar lowers the horizontal base line ; in Fig. 1 the fall corresponds to a volume change of no more than 1 c.c., but it may be greater than this depending on how the arm is arranged and the grasp made. The lowered base line remains horizontal while contraction is maintained uniform, returning to its original level when the muscles are relaxed. When the circulation to the arm is free, the volume changes caused by contraction and relaxation are greater.

It is clear that the method is suitable for the measurement of blood flow not only when the arm is at rest before and after exercise but also during sustained muscular contraction. Because of the recurring oscillations it is not well adapted for measurement during rapid rhythmic contractions,

Fig. 1. Effect of muscular contraction and relaxation on forearm volume in an arm depleted of blood. In the right hand tracing the movements were made more slowly than in the left. (D.S., 7.10.37.)

In this and subsequent figures, the duration of the contraction is indicated by horizontal white lines ; calibration in c.c. on right ; time marker in 10 seconds beneath.

though rough estimates of blood flow can then be made from the general slope of the series of oscillations when the congesting pressure is applied to the upper arm. These estimates, however, are of doubtful value. In what follows, blood flow rates are stated in c.c. of blood per minute per 100 c.c. of limb volume enclosed within the plethysmograph. The times at which blood flows are stated to be measured are those of the moment when congesting pressure is applied to the upper arm and indicate the flow during the next 5 or 10 seconds ; the congesting pressure is maintained for this period to allow inscription of a length of curve sufficient for measurement. If required, successive inflow curves can be inscribed at intervals of 15 or 20 seconds, 5 to 10 seconds being allowed to elapse between the release of congesting pressure and its next application to permit the collected blood to escape from the veins. Reliable curves may be obtained within a few seconds of the beginning of contraction or after the end of exercise, when the

oscillations due to muscular movement have subsided. The correct and rapid manipulation of the apparatus requires practice.

Resting blood flow.

It was previously shown (11) that after about a half hour's rest (to allow the effect of previous use to subside) the blood flow rate in the lean and muscular forearm of a subject maintained comfortably warm lies between 1 and 2 c.c. (average about 1·5 c.c.).* It varies but little from subject to subject or in the same subject at different times and, in contrast to the hand, is not greatly increased or lowered by warming or cooling the body. It seems considerably smaller and much more constant than that obtained in animal experiment, though the data available for comparison are scanty ; few workers state flow in units of muscle as well as of blood and time. For example, and interpreted into the blood flow rate now used† the resting rate in the levator muscle of the lip of the normal horse (Chauveau and Kaufmann (5)) varies between 6·7 and 37·4 c.c. (average about 19 c.c.), in the gastrocnemius and digastric muscles of the anæsthetised dog (Barcroft and Kato (4)) between 1·4 and 44 (average, from their Table VI, about 17 c.c.), and in the gastrocnemius of the anæsthetised cat (Verzár (16)) between 3·4 and 32 c.c. (average, from his Table I, about 12 c.c.). It will be seen later that the level of the resting blood flow is of importance in dealing with the effects of sustained muscular contraction.

Effects of Exercise.

Many previous workers, using various methods on animals, find that muscular contraction mechanically compresses the vessels and that during a sustained contraction blood flow through the muscles is greatly diminished and may even cease ; according to Anrep and others (1 and 2) flow is diminished in proportion to the strength of contraction. Gaskell (9 and 10) states, however, that if the contraction lasts some time, blood flow may increase while the muscle remains contracted. Keller, Loeser and Rein (13) find that, after an initial and transient increase, blood flow during a sustained contraction remains either at the resting level or gradually and progressively increases ; they believe that muscular contraction favours blood flow by

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* Further rest usually reduces the rate slightly, to about 1 c.c.. This reduction comes in spite of repeated exercise of the forearm and is independent of the sympathetic nerves. For example, in a normal subject the blood flow rate was 1·4 c.c. after 30 minutes' rest. During the next 2 hours, the arm being kept at 30°C. in a water bath, the resting rate fell to 0·9 c.c. although the arm was exercised at intervals during this time. Again, in one patient whose right arm was deprived of the sympathetic nerves by cervical ganglionectomy six months previously, the blood flow rate showed a similar fall (from 2·1 to 1·8 c.c.) with prolonged rest. The cause of this fall is unknown but it is thought to be due to a circulating adrenaline like substance which is reduced in concentration by prolonged rest.

† Chauveau and Kaufmann (5) state blood flow in grammes and Barcroft and Kato (4) and Verzár (16) in c.c. of blood per gramme of muscle per minute. Grammes and c.c. of blood and muscle are approximately equivalent, their specific gravities being but little over 1000.

reducing resistance. Frey (8) also finds a progressive dilatation of arteries and veins of all calibres beginning a few seconds after the onset of a sustained contraction. All who deal with the after effects of contraction are agreed that with relaxation there is an immediate and conspicuous increase of flow followed by a gradual return to the previous level. The new observations in man provide evidence supporting the general conclusions but they show that although contraction does compress the vessels, the compression is insufficient to prevent a dilatation and increase of flow up to a certain level while contraction is maintained. They also suggest a means of reconciling the conflicting views.
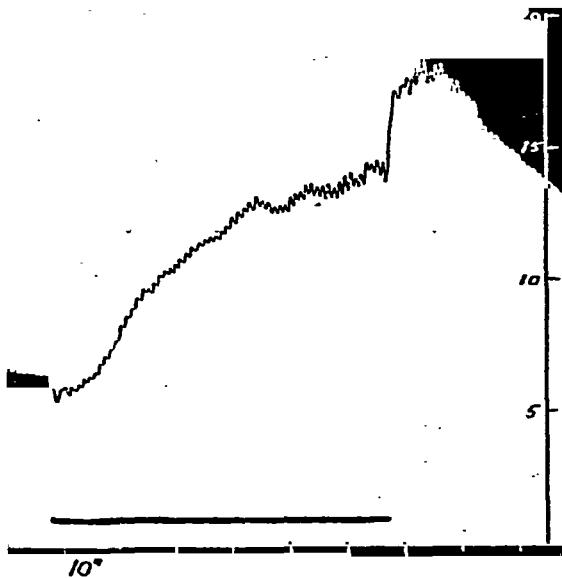
Fig. 2. Effect of strong contraction, sustained for about a minute, on forearm volume. (D.S., 11.10.37.)

Effects of a single sustained contraction. When the bar is strongly grasped and the grasp is maintained at its full strength,* limb volume at first falls (Fig. 2). The fall is greater than that happening when the limb is depleted of blood; the increase is interpreted as due to blood being squeezed out of the forearm by the contracting muscles. Gaskell (9) and others have observed an outrush of blood from the veins when a muscle contracts. After 3 to 5 seconds, however, the base line begins to rise and

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* It can be so maintained for 1 to 2 minutes.

rises considerably to reach a maximum, after about 30 seconds, which is maintained so long as the contraction lasts with undiminished force. The pulse beat also increases greatly. When the grasp is released there is at once a further but smaller rise of volume and increase of pulse beat ; these subside gradually to the resting level. Blood flow measurements show that almost immediately following contraction, the rate begins to increase rapidly and, after half to one minute reaches a maximum of 4 to 5 c.c. which is maintained for the duration of the grasp. The interval between the subsidence of the oscillations of the recorder, due to contraction, and the beginning of the rise is usually too short to allow blood flow to be measured then. On occasions when it can be measured, the resting flow is found to be unaltered or slightly reduced. Immediately after relaxation, the flow increases enormously and thereafter declines, at first rapidly and then more slowly, to the former resting level. With gentler grasps smaller effects are obtained. A typical example is given in Fig. 3.
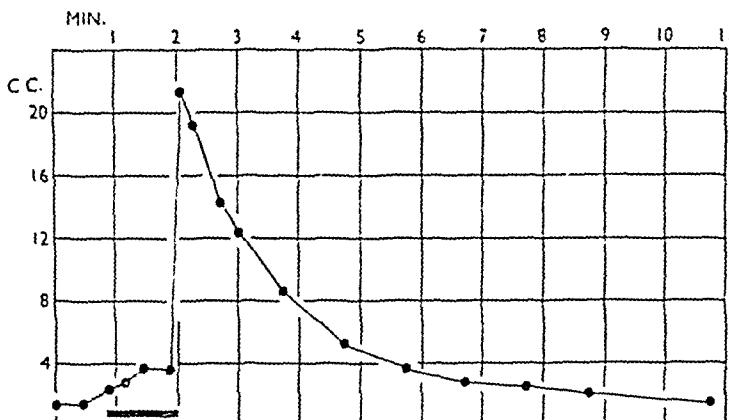
Fig. 3. Forearm blood flow rate before, during and after a strong contraction maintained for 1¼ minutes. (A.B., 29.10.37.)

These results can be obtained with but little variation from different subjects or from the same subject at different times under standard conditions. They indicate that while strong muscular contraction does compress the vessels, this compression is insufficient to prevent them dilating to a certain extent (under the influence of dilator substances released from the muscle fibres—see later for this point). The compression is removed with relaxation. Rein (13) and his colleagues, however, interpret similar observations in the dog by supposing that contraction does not compress the vessels but rather favours dilatation and that dilator substances are not set free from the muscle fibres until they relax. There is strong evidence against these suppositions.

In the first place if the arm is exercised sufficiently to provoke a large increase of limb volume and blood flow, then a strong sustained contraction immediately reduces limb volume considerably and lowers blood flow

greatly; these reductions persist while the contraction is maintained; high levels are at once regained on relaxation. Fig. 4 shows the reduction of limb volume caused by three sustained contractions of an arm previously exercised. Moreover, under these conditions the high blood flow following exercise is reduced by the sustained contraction to about the maximum level attained during a long lasting contraction in the previously resting arm. For example, in the same subject as in Fig. 3 and following the observation there illustrated, the forearm muscles were exercised by grasping and releasing the bar once a second for 2 minutes. Immediately after cessation of the exercise the blood flow rate was 35·5 c.c.. The bar was then grasped firmly for half a minute and during this time blood flow was measured
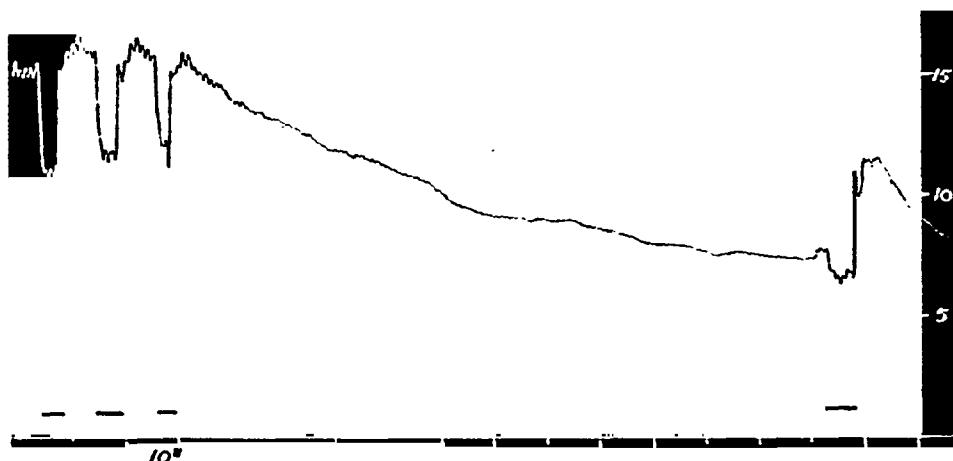
Fig. 4. Forearm volume record showing, on left, effect of 3 successive contractions of previously exercised muscles, and on right another contraction made after limb volume had almost returned to resting level. (D.S., 11.10.37.)

twice, being 4·5 and 4·0 c.c.. The grasp was released and blood flow returned immediately to 32·0 c.c.. Secondly, if the forearm blood flow is increased by means other than exercise, by arresting the circulation, then a strong contraction of the muscles again reduces blood flow greatly. For example, in one subject the blood flow following arrest of the circulation for two minutes was found to be constantly about 20 c.c., but if the bar was firmly grasped just before, the grasp and maintained for a half minute after the circulation was restored to the arm, blood flow was no more than about 5 c.c. while the grasp was maintained (4·4 to 6·4 in separate observations). Blood flow rose when the grasp was released. A mechanical compression of the vessels during muscular contractions seems a reasonable conclusion from these findings. As Anrep (1) and others find in the dog, so in man,

develop. The observations suggest another possible reason why some workers have failed to observe vasodilatation during sustained contraction. In animal experiment the resting blood flow is much higher than in the normal man ; if the " resting " blood flow is already so great as to be beyond that which can develop during a tetanus then only a reduction of blood flow is to be expected when the muscle contracts.

Effects of repeated contractions. It has already been seen that in the resting arm with narrow vessels, a strong contraction of short duration of the forearm muscles causes at first only a small reduction of limb volume and little or no change in blood flow. When such a contraction is released,
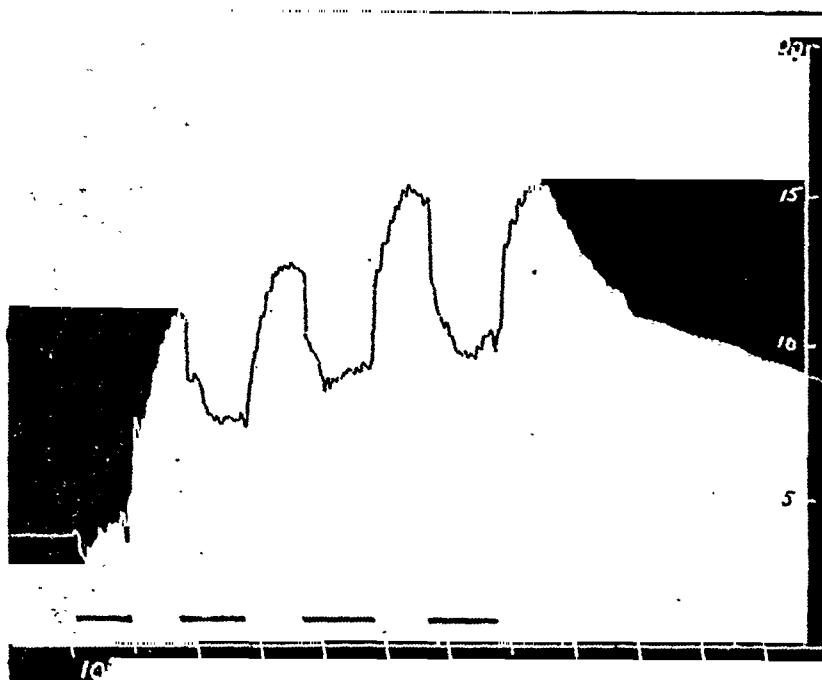
Fig. 6. Effect on forearm volume of 4 successive contractions, each maintained for about 10 secs.. (D.S., 11.10.37.)

there is an immediate and considerable increase in volume and flow (Fig. 4, right hand contraction). If other similar contractions are made in succession, each at the moment when the after effects of the previous one is at its height (namely, an interval of about 10 seconds between relaxation and contraction) then as is shown in Fig. 6, the reduction in limb volume with each contraction increases rapidly to reach a maximum. Limb volumes during both contraction and relaxation soon rise to and maintain high levels ; these are but little raised by further exercise. Blood flows measured during successive contractions and relaxations show that a maximum of 5 c.c. is rapidly attained from contracted muscle while during relaxations blood flow rises at first rapidly and then more slowly towards a maximum of between 25 and

35 c.c.. From this it can be estimated that the average blood flow rate during such an exercise after the maximum has been attained is between 15 and 20 c.c.. It has already been mentioned that the method is unsuitable for estimating blood flow during more rapid rhythmic exercise but it is clear that the rate must lie between the maxima attainable in the phases of contraction and relaxation and must depend on the relative durations of contraction and relaxation. According to Chauveau and Kaufmann (5), the blood flow rate attained in the horse while chewing oats varies from 56·5 to 123·5 c.c. (the average being about 76, or about four times the resting flow). In Barcroft and Kato's (4) observations in which a muscle of the dog's leg was stimulated electrically once a second for 15 minutes the rates are even more variable, form 2·1 to 110 (average from their Table VI being about 30, or hardly double the average resting value).

Fig. 7. Effect of two quick, short and strong contractions. Note the large oscillations due to contraction and relaxation; also the normal variations of limb volume returning after (first contraction) and before (second) the increase of limb volume following contraction subsides. (D.S., 10.9.37.)

In dealing with after effects of repeated contractions, a standard exercise has been used throughout, namely, a short strong grasp of the bar lasting about a half second and repeated every second. The exercise is easily performed by unpractised subjects and most of them can continue for 4 to 5 minutes before the forearm muscles are fatigued. It is not feasible to ensure that the exercise is always carried out in exactly the same way as regards strength and duration of the contractions but even so the results are in good agreement in different subjects and in the same subject from time to time. The end point of the hyperæmia following exercise is often not precisely determinable because the small variations of resting blood flow, indistinguishable during the height of the hyperæmia reappear as this subsides (Fig. 7) sometimes in an exaggerated form.

Immediately after exercise limb and pulse volume and blood flow are greatly increased; they subside at first rapidly and then more slowly to the resting level. In general, it can be said that the longer and the more strenuous the exercise the greater and the longer lasting the after effects. After 4 or 5 minutes of exercise the blood flow rate reaches 25 to 35 c.c. and

does not return to resting level for 15 to 30 minutes. These findings, exemplified in Table I, are in general agreement with previous work, though comparable data are lacking. Even with longer exercise, using maximal contractions, the hyperæmia is neither materially increased nor greatly prolonged beyond these limits. Barcroft and Kato (4) record a hyperæmia lasting several hours (in their Experiment 11 over seven hours) after 15 minutes' rhythmic stimulation of a dog's muscle; nothing comparable to this has been observed in the human limb.

TABLE I (Normal Subject).

Duration of exercise.	Resting rate before exercise.	Immo-diately after exercise.	Blood flow rates in c.c. per 100 c.c. limb volume per min..												
			Minutes later.												
			$\frac{1}{3}$	$\frac{2}{3}$	1	$1\frac{1}{2}$	2	3	4	5	7	9	11	13	15
1 sec.	1·4	11·1	3·7	1·7	1·4										
5 secs.	1·4	12·6	6·9	5·4	2·8	3·2	1·6	1·4							
20 secs.	1·1	19·5	15·7	9·2	7·7	3·2	2·3	2·8	2·0	1·9	1·7	1·1			
40 secs.	0·9	29·2	24·0	15·5	10·9	5·7	2·8	2·0	2·8	2·0	1·9	1·7	1·1	1·0	
4 mins.	0·9	33·2	25·7	16·9	12·9	7·5	6·0	3·4	2·3	2·3	1·4	1·3	1·1	1·1	0·9

D.S., 16.11.37. Water bath 30°C.. Room temperature 13°C.. Limb volume 700 c.c.. Subject comfortably warm.

The mechanism of the vasodilatation due to exercise.

It is generally accepted that, as Gaskell (10) first suggested, the increase of blood flow brought about by exercise is due to the release of vasodilator substances from the active muscle fibres and that vasomotor nerves play little if any part. Observations on man bring fresh evidence on both these points.

(a) *Sympathetic nerves.* Our knowledge of the distribution and function of the sympathetic nerves to voluntary muscle is still uncertain and, so far as I am aware, no data are recorded as to the effect of exercise on muscle blood flow in limbs after section and degeneration of the sympathetic nerves. The observations on sustained and repeated contractions described in the foregoing pages have been repeated on one patient six months after right cervical ganglionectomy for relief of symptoms due to arterial obstruction in the right

TABLE II.

Comparing two arms after right sided sympathectomy.

Blood flow rates in c.c. per 100 c.c. limb volume per minute.

Duration of exercise	Fore-arm	Before exercise	Immediately after	Minutes after exercise														
				¼	½	¾	1	1½	2	3	4	5	7	9	11	13	15	23
1 sec.	L.	1·3	11·4	3·3	3·3	3·0	2·5	1·8	1·5	—	—	—	—	—	—	—	—	—
	R.	2·1	15·6	5·2	3·0	2·5	2·1	—	1·5	—	—	—	—	—	—	—	—	—
5 secs.	L.	1·5	13·2	4·5	4·2	3·9	2·7	1·8	1·5	—	—	—	—	—	—	—	—	—
	R.	1·8	18·0	8·7	7·5	4·0	3·0	2·2	2·1	1·8	—	—	—	—	—	—	—	—
20 secs.	L.	1·6	13·8	12·3	11·7	6·9	5·7	3·6	3·0	2·7	2·2	2·1	1·8	—	—	—	—	—
	R.	2·1	20·7	17·4	9·0	7·8	6·2	4·0	3·3	3·0	2·5	2·2	2·1	—	—	—	—	—
40 secs.	L.	1·2	18·6	17·1	11·7	8·7	7·8	4·8	3·3	3·0	3·0	2·7	2·1	1·5	1·2	—	—	—
	R.	2·1	20·4	19·8	15·3	12·3	7·8	6·0	5·4	4·2	3·7	3·6	3·0	2·7	2·4	2·1	—	—
4 mins.	L.	1·2	24·0	20·1	17·4	15·0	13·2	11·1	10·5	8·4	7·5	5·7	3·0	2·1	1·8	1·2	—	—
	R.	1·8	22·2	21·9	15·6	8·7	7·2	4·8	3·9	3·6	3·9	4·2	3·6	3·6	2·7	2·1	2·1	1·8
5 mins. inclusive	L.	1·2	37·2	20·4	11·7	5·7	3·0	2·1	1·8	1·8	1·3	—	—	—	—	—	—	—
	R.	1·8	40·8	20·1	17·1	9·0	6·3	3·0	3·6	3·3	3·3	3·3	2·4	2·4	—	—	—	—

S.K., 11.11.37. R. and L. limb volume 500 c.c. Bath temperature 30°C. Room temperature 14°C. Subject comfortably warm.

hand.* The results, exemplified in Table II, reveal no essential difference between the hyperæmia of exercise in the right arm long deprived of its sympathetic nerves and that in the left normal arm.   A minor difference is that while the maximum hyperæmia attained by long exercise is about the same in the two arms, the hyperæmia provoked by shorter exercise is greater in the right than in the left arm.   This is probably due to the increased sensitivity of denervated vessels to stimuli of all kinds.   The greater dilator effect of minute doses of adrenaline in this patient's right arm as compared with his left was described in a previous paper (11).   Circulatory arrest also provokes a greater and longer lasting hyperæmia in the right than
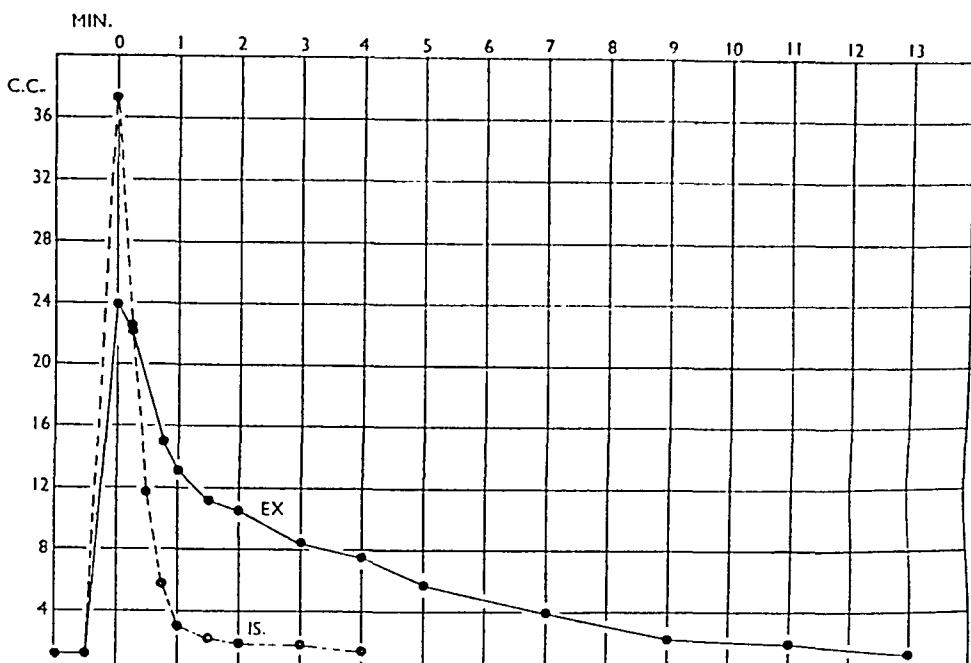
Fig. 8.   Forearm blood flow rate before and after four minutes' standard exercise (continuous line) and five minutes' ischæmia (broken line).   Time in minutes after end of exercise and after restoration of circulation.   (S.K., 11.11.37.)

in the left forearm (Table II).   It may be concluded, therefore, that the sympathetic nerves play no essential part in provoking the local vasodilatation of exercise.

(b) *Dilator substances.*   Gaskell (10) drew attention to the resemblance of the effects of sustained contraction to those of circulatory arrest and he considered the possibility that the hyperæmia following a contraction might be due to anæmia caused by the contracted mass of the muscle.   He found, however, that the effects of a short tetanus are greater than those of a longer period of ischæmia.   It has been seen that in man, while there is a

* This patient is referred to in the previous paper (11).   The circulation to both forearms was normal.

compression of the vessels by contracted muscle, the compression is insufficient to prevent them dilating so that ischæmia during contraction cannot be regarded as the cause of the hyperæmia following exercise. There is further evidence for this view. In man the chief difference between the hyperæmia of exercise and that following a similar period of circulatory arrest is the greater duration of the former, (see also Lewis and Grant (14)). As is shown by the example given in Fig. 8 following 5 minutes circulatory arrest, the blood flow rate rises to 37·2 c.c. but this largely subsides within a minute and returns to the previous level within 4 minutes. After 4 minutes of the standard exercise of grasping the bar, the blood flow of 24 c.c. subsides much more slowly, and returns to resting level only after 13 minutes. Though the degree of hyperæmia following ischæmia is apparently greater than that after exercise it is to be remembered that in the former the hyperæmia affects all the tissues of the forearm while in the latter it affects only or mainly those of the active muscles; if allowance is made for this the hyperæmias are of the same order of intensity; that following arrest is
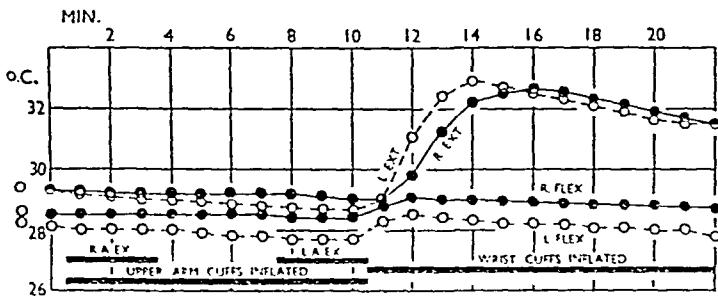
Fig. 9. Skin temperatures on flexor and extensor aspects of both forearms (R.L. Ext. and Flex.). Durations of exercise and of inflation of arm and wrist cuffs indicated by horizontal lines. (R.G., 27.10.37.)

strikingly more transient than that after exercise. This difference is reflected also in skin temperature, which rises but little after circulatory arrest; if the muscles are exercised during a period of ischæmia skin temperature rises conspicuously when the circulation is restored (but not until then) (Grant and Pearson (11)).

That metabolites are responsible for vasodilatation in various organs has been amply confirmed since Gaskell's time, but there is little direct evidence for voluntary muscle (see Barcroft and Kato (4)). Additional evidence is provided by showing that the hyperæmia of exercise is delayed by circulatory arrest, as is exemplified by the following experiment. Both forearms are exposed, pronated and resting on sandbags at the elbow and wrist, sphygmomanometer cuffs being applied to both upper arms, and to both wrists, but not inflated. Skin temperatures are recorded from the

flexor and extensor aspects of both (Fig. 9). When the skin temperatures
are sufficiently low the circulation to both upper arms is arrested. The
right wrist is extended and dropped once a second for three minutes and then
allowed to rest.* Four minutes later the left extensor muscles are exercised
in the same way and at the end of three minutes, both wrist cuffs are inflated
and the upper arm cuffs released. The skin flushes brightly and returns to
normal after three minutes. The temperatures of the flexor aspects show
only the small rise due to the reactive hyperæmia. Those of the extensor
aspects both rise quickly and rise at the same time to equal heights and fall
simultaneously, although the exercise of the right arm finished seven minutes
before that of the left. Again as is shown in Fig. 10, in patients suffering
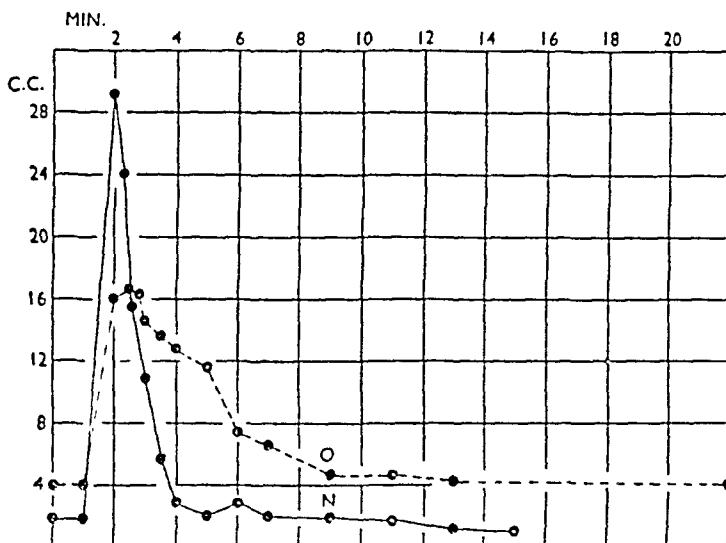
Fig. 10. Forearm blood flow rate before and after 40 secs. standard exercise in a normal subject
(continuous line) and in a patient (broken line) suffering from thromboangeitis obliterans,
the brachial artery being blocked above the elbow. Note the unusually high resting rate
in the patient, due probably to a slight tremor of the muscles. (D.S., 16.11.37 ; H.P.
8.11.37.)

from arterial obstruction† the hyperæmia of exercise (like reactive hyperæmia)
though reduced in degree, is prolonged in time. These observations point
strongly to relatively stable metabolites as being responsible for the vaso-
dilatation due to exercise.

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* This exercise brings into play the extensors but avoids synergistic contraction of the
flexors.

† It has been pointed out (11) that Hewlett and Zwaluwenburgs' (12) method is not always
suitable for measuring blood flow in patients in whom the main arteries of the limb are blocked,
owing to compression of collateral vessels. If, however, as the case illustrated in Fig. 10, the
blood flow rate is not increased by reducing the congesting pressure from 60, in 10 mm. steps,
to 30 mm. Hg, it may be concluded that the arteries supplying the forearm are not being
compressed.

SUMMARY AND CONCLUSIONS.

1. Using mainly a plethysmographic method, observations are described on the local vascular effects of exercise of the voluntary muscle of the human forearm.

2. The resting blood flow rate is much lower and more constant than is found in animal experiment.

3. A sustained contraction of the muscles compresses the vessels; the degree of compression depends on the strength of contraction; it is insufficient to prevent, but controls the degree of, dilatation and increase of flow which develop while contraction is maintained. The compression is removed with relaxation.

4. After exercise of the muscles, blood flow is greatly increased; the longer and the more strenuous the exercise the greater and the longer lasting the subsequent hyperæmia.

5. The local vascular effects of exercise are independent of the sympathetic nerves.

6. Evidence points strongly to relatively stable metabolites as being responsible for the hyperæmia of exercise.

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OBSERVATIONS ON REFERRED PAIN ARISING FROM MUSCLE.

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LEWIS has recently shown (4) that the quality of pain provoked from somatic structures depends more upon the structure stimulated than upon the nature of the stimulus. To provoke pain he has injected small quantities of various substances into structures such as muscle, tendon, and periosteum, and has found that each structure gives rise to pain which is characteristic, but independent of the substance injected. In particular he has found that the pain produced by injecting such substances into muscle is identical with that resulting from muscle contracting under ischæmic conditions, and therefore represents true muscle pain.

During these experiments Sir Thomas Lewis noticed that pain arising from muscle might be referred to a distance, and he suggested that, using the injection method, I should make in his laboratory a systematic examination of pain provoked from the accessible muscles of normal subjects.

Clinical observers (3, 5, 6) have pointed out that painful conditions of the extremities may be associated with tender areas in the muscles of the limb girdles, and have suggested the possibility that pain arising from muscle may be referred. The following observations were carried out as a preliminary study, with a view to subsequently exploring the occurrence and possible significance of pain referred from muscles in patients.

Method.

The injection of chemical solutions is a very convenient method of producing pain from any muscle or any given part of it. Hypertonic saline was chosen as the most satisfactory agent. A 6% solution of sodium chloride when injected into muscle in amounts of 0·1 to 0·3 c.c. gives rise to continuous pain, which rises rapidly to a maximum of considerable severity, and subsides more slowly over a period of 3 to 5 minutes. It leaves no undesirable after effects.

Different muscles were found to differ considerably in sensitivity. For instance 0·1 c.c. 6% saline when injected into the rectus abdominis or into

the small muscles of the hand gives very severe pain, while the same quantity injected into the biceps or glutei gives only slight pain. For this reason different stimuli were used on different muscles, the amounts varying from 0·1 c.c. of 4% to 0·3 c.c. of 6% saline.

Injections of similar quantities of 1% methylene blue into cadavers showed that the dye could be accurately placed inside muscles and did not spread widely through the tissues. The portion of muscle coloured by the larger quantities of dye was about 2 cm. × 3 cm. × 1 cm. though the exact distribution of the dye varied considerably with the different arrangements of muscular fibres in different muscles. On rare occasions injected saline would seem to have stimulated nerve trunks. These have been small nerves such as the intercostal or digital nerves, and the result has been a sensation of burning pain and pins and needles felt in the cutaneous distribution of the nerve in question. This sensation is strikingly different from that produced by saline injected into muscle, and the two are not likely to be confused.

The observations have been carried out chiefly upon myself and other workers in the laboratory. In many of the critical experiments I have had the good fortune to obtain the help of colleagues who are themselves working on other aspects of sensory disturbances, and are unusually experienced in subjective observation.

No statement is made about the distribution of pain which has not been observed by at least three subjects.

Differentiation of muscle and fascial pain, and constancy of reference.

A muscle may be considered as being made up of three parts, a fleshy belly, tendon, and fascial covering and tendon sheath. Referred " muscular " pain might arise from any of these structures. To determine which structure is responsible two sets of observations were made.

For the first the gluteus medius was chosen because of the tough well developed fascia which covers it. Three buttons of novocaine were introduced into the skin of the buttock 4 cm. apart. Three large intramuscular needles were passed through the anæsthetised skin, and were moved about in the subcutaneous tissue without any pain being appreciated. The needles were then impinged in turn on the gluteal fascia, which could be felt easily as a resistant layer. The subject was unaware of which needle was being moved. Stimulation of the fascia in this way produced pain which was recognised as arising from a point localised regularly a few centimetres distal to the needle concerned. 0·1 c.c. of 6% saline was then injected through each needle into the fascia. This gave a similar pain localised in the same way. This " fascial " pain was easily distinguished from " skin " pain by its different quality. The needles were then passed deeply into the muscle. Moving the needle point about vigorously gave rise to a very slight diffuse pain felt in most of the buttock. 0·2 c.c. of 6% saline was then injected into the muscle. This gave a diffuse pain of

greater severity felt clearly in the lower part of the buttock and the back of the thigh, and occasionally as far distant as the knee. These observations were repeated on two other subjects in exactly the same way and with similar results. Pain derived from the fascia is confined to the neighbourhood of the point stimulated, and pain from the muscle is felt diffusely and is referred down the back of the thigh (Fig. 1).
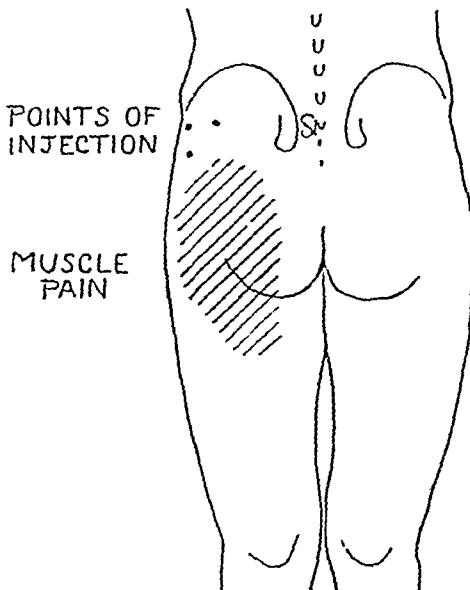

Fig. 1. Hatched area represents the distribution of slight pain from gluteus medius stimulated at points of injection.

The second set of observations was repeated on 14 different subjects, the object being to see if the reference is constant in different individuals, and if it changes when different parts of the same muscle are stimulated. For this the tibialis anticus was chosen because of its accessibility and simple structure. The procedure was similar. Three buttons of novocaine were introduced into the skin over the tibialis anticus ; the first and second over the upper and middle parts of its fleshy belly (about 8 cm. apart), and the third over the tendon where it lies in front of the ankle. Hypodermic needles were then passed through each anæsthetised patch of skin and made to scratch the fascia or tendon sheath. This produced severe pain which was always recognised as coming from a point localised regularly about 2 cm. distal to the needle by most subjects and accurately at the needle by a few. The needle was then passed deeply into the muscle at the uppermost point, its introduction giving no pain after the fascia had been pierced. 0·1 c.c. of 6% saline was then injected into the muscle. This gave a diffuse pain felt by most subjects in front of the ankle and in the outer and middle part of

the front of the leg.   A few subjects had ankle pain only, and one subject had pain only in the leg.   The results obtained from injection at the second point of the muscle were similar.   Passing the needle into the tendon occasionally gave a purely local pain.   The injection of 0·05 c.c. of 6% saline into the tendon substance gave diffuse pain felt over a small area on the medial aspect of the instep, this result being uniform in all subjects.   At first saline was also injected into the fascia and tendon sheath, but as the pain produced did not differ from that produced by passing the needle this was discontinued after the first few subjects (Fig. 2).
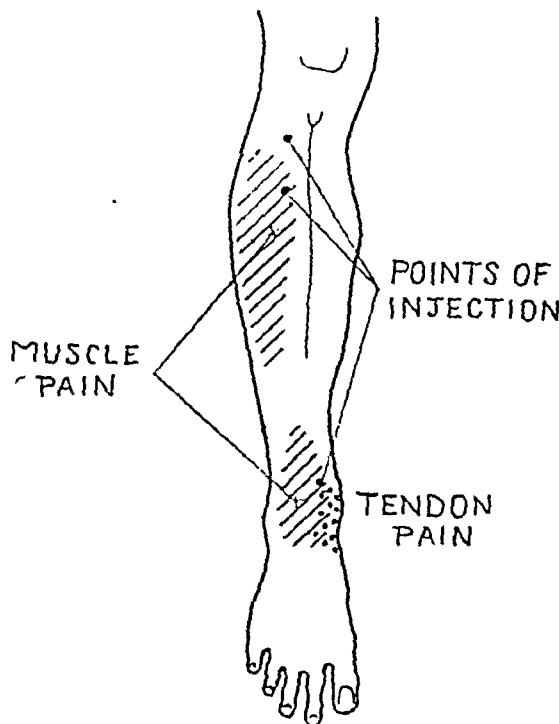
Fig. 2.   Distribution of pain from tibialis anticus stimulated at points of injection :   muscle pain hatched :   tendon pain stippled.

It appears from these observations that the gluteal and anterior crucal fasciæ are sensitive structures, and that pain arising in them can be recognised as coming from points which can be localised with fair accuracy.   The muscle bellies are less sensitive, and in them a local stimulus is not recognised as such, pain being felt diffusely over a considerable area which may be situated at a distance from the point stimulated.   Pain from tendon is intermediate between these two.   With a given stimulus pain from the tibialis anticus is always referred in the same general way ; though there is of course some variation in the distribution from individual to individual. There is also some variation with the point of muscle stimulated ; thus, the higher point gives pain in the leg at a higher level and ankle pain more

frequently than the lower. With muscles of more complex structure the reference is more variable. For instance in the triceps the distribution of the pain varies according to which head of the muscle is stimulated, and the pain may be referred to almost any part of the upper extremity.

Local pain from muscle.

In certain muscles the injection of saline gives rise to a sharply localised pain in addition to the more diffuse pain which may be referred. This very local pain is similar to the pain derived from fascia. The trapezius, pectoralis major, deltoid, serratus magnus, vastus internus are examples of muscles giving rise to clearly defined local pain ; this local pain is, however, present to a lesser extent in the majority of muscles. It will be noticed that these muscles form superficial sheets permeated by fascial septa, and it seems probable that in them the saline stimulates both muscular and fascial elements. There would seem to be a difference between this sharply localised pain, and the diffuse muscular pain even when the point stimulated lies within the distribution of the latter.

Some examples of reference.

Varying amounts of hypertonic saline were injected into a large number of muscles in different individuals and the character and distribution of the referred pain noted. Some general characteristics of these pains will be described first.

As already stated the pain is felt diffusely over a wide area, often situated at some distance from the point stimulated. The extent of the area increases with the strength of the stimulus. The smallest area seemed about 4 cm. square in extent, lesser stimuli being unappreciated, or felt as a vague discomfort in the same region. The pain is not distributed evenly, there being usually one or more small regions of maximal intensity surrounded by a zone of slight pain. When there are two or more regions of maximal pain they are rarely appreciated simultaneously, the pain being felt most in the several regions successively. This phenomenon is sometimes described by subjects as a " moving " or " shooting " pain.

Some of these regions of maximal pain are described as situated in structures other than muscle. In the limbs the pain is often described as felt deeply near joints. For instance 0·1 c.c. of 6% saline injected into the muscle of the infraspinous fossa, gives pain felt deeply at the shoulder tip. Similarly pain from the peroneus longus is described as felt in the ankle and pain from the vastus intermedius in the knee. Pain from the flexor digitorum profundus, may seem to come from the knuckles ; and when this is compared with the pain produced by injecting 0·3 c.c. of 6% saline directly into the joint space of the corresponding metacarpo-phalangeal joint in the opposite hand, the two pains are stated to be very similar if not identical. From these examples it is clear that pain arising in muscle and referred to

the region of a joint may easily be confused with pain arising in the joint itself.

In the head, other structures may be involved. 0·1 c.c. of 6% saline injected into the masseter where it lies over the angle of the mandible produces pain felt mostly within the mouth in the region of the upper jaw, which is described as " toothache " ; some pain may also be felt in the region of the temperomandibular joint, and the external auditory meatus. The same quantity of saline injected into the small muscles of the sub-occipital triangle gives pain felt deeply in the head which is described as " head ache," and saline injected into the occipitalis or the facial muscles in the region of the canine fossa may give rise to " ear ache," though in the face the saline produces much " skin " and " facial " pain which makes the more diffuse pain difficult to recognise.

The back muscles also give interesting examples of reference. 0·2 c.c. of 6% saline injected into the superficial layers of the erector spinæ in the mid-lumbar region gives pain felt over the upper part of the buttock ; this pain feels more superficial than most muscular pains and when it is compared with the pain produced by injecting saline into the gluteal fascia of the opposite side the two pains are found to be very similar in quality. The same quantity of saline injected deeply into the multifidus opposite the 1st and 2nd lumbar spines gives less pain in the back, and may give pain felt in the region of the scrotum of the same side, which is very similar to pain produced by squeezing the testis of the opposite side.

From these observations it would seem that pain arising from muscle may often fail to be recognised as such, and may be ascribed by the subject to other structures such as joints, teeth, or testis.

Segmental distribution.

In order to decide if the distribution of referred muscular pain follows a spinal segmental pattern, it becomes necessary to record the distribution of the pain with accuracy. This is not always easy. Firstly, the pain is diffuse. Secondly, there is individual variation, in that some subjects describe only the distribution of the regions of maximal pain while others give the complete zone of slight pain as well. Thirdly, the distribution varies with the severity of the pain. But if subjects displaying full areas of distribution are used, and if the stimulus is graded to give pains of comparable severity, and the subject repeatedly marks out the limits of the pain while it lasts these difficulties largely disappear.

Trunk. The trunk muscles were investigated first because of their simple segmental innervation. Saline was injected at three points ; into the 9th intercostal space in the mid-axillary line, into the multifidus opposite the 9th thoracic spine, and into the rectus abdominis 3 cm. above the umbilicus ; the presumption is that the muscle at these three points is supplied by the 9th thoracic nerve. The rectus gave severe pain in front in the hypochondrium and slight pain in the back. The multifidus gave

severe pain in the back and slight pain in front; while the intercostal space gave moderate pain both front and back. The regions in which pain was felt were the same in these three observations, only the relative intensity varied as between front and back (Fig. 3). Similar observations were made at different levels from the second intercostal space to the pubes, and were
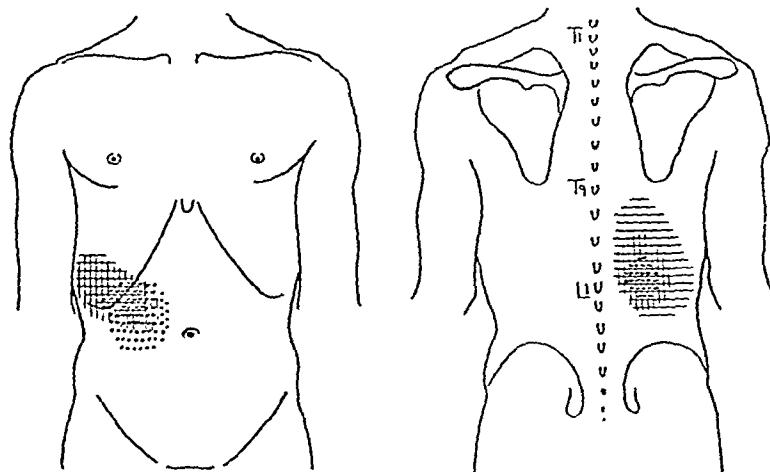

Fig. 3. Distribution of pain from muscle supplied by 9th thoracic nerve; when multifidus injected horizontal hatching; when intercostals injected vertical hatching; when rectus abdominis injected stippling.

repeated on different subjects. The results were substantially the same except that at certain levels the erector spinæ gave no pain in front, and that in some subjects the reference from back to front was unobtainable.

Three neighbouring intercostal spaces were injected and the resulting pain areas were found to be situated one above the other at levels differing by the width of a rib and intercostal space (Fig. 4).
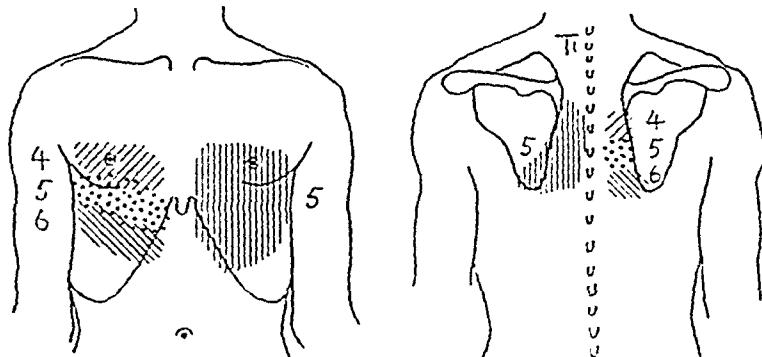
Fig. 4. Distribution of pain from intercostals. Right side shows slight pains from 4th, 5th and 6th spaces. Left side shows very severe pain from 5th space.

It will be noticed that the pain areas together cover most of the limb and present little overlap.

Muscles with motor supply from more than one segment were chosen, for instance,

Infraspinatus $C_{5, 6}$. Serratus anterior $C_{5, 6, 7}$. Latissimus dorsi $C_{6, 7, 8}$. The distribution of the referred pain from these muscles is shown in Fig. 7.
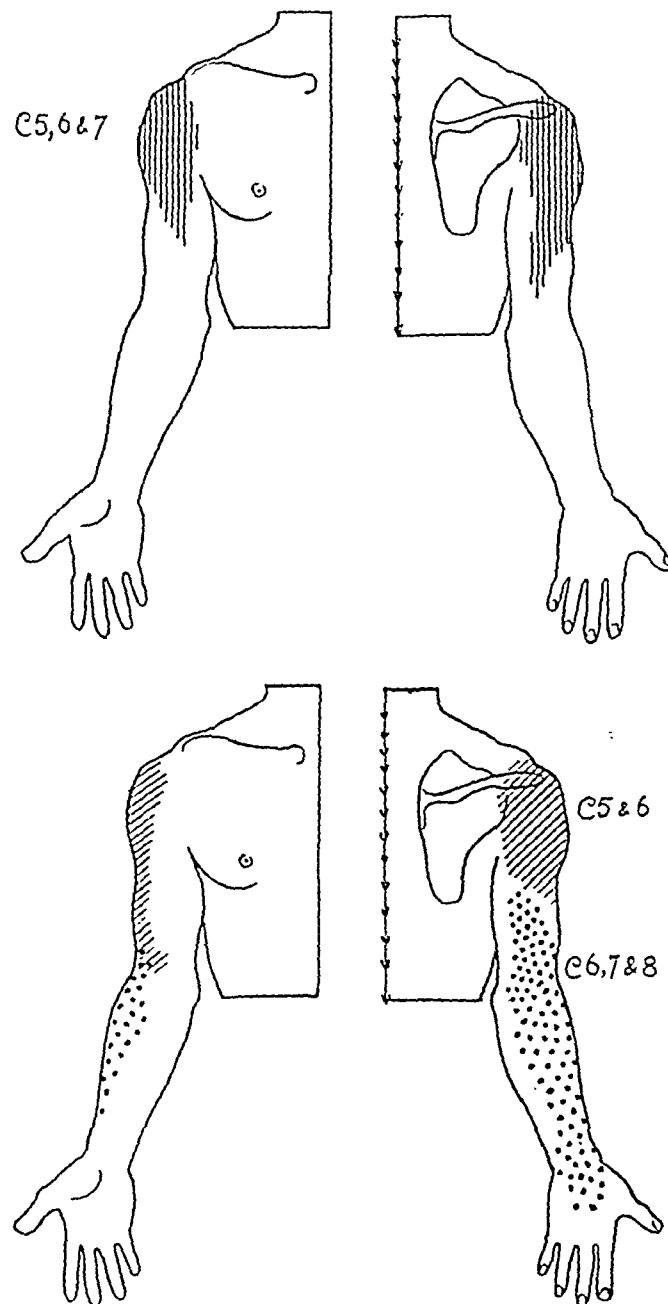

Fig. 7. Distribution of referred pain from arm muscles. From serratus anterior vertical hatching; from infraspinatus oblique hatching; from latissimus dorsi stippling.

It will be seen that the pain areas fall within the corresponding segmental areas of reference obtained by the previous experiment. Other muscles of the arm when tested gave referred pain distributed similarly in accordance with expectation.

In the leg there are no muscles supplied on the motor side from a single segment, and an analysis as simple as in the case of the arm cannot be accomplished. Injections into a large number of muscles have shown the pains to be referred widely up and down the leg ; broadly speaking, the muscles supplied by the 2nd, 3rd, and 4th lumbar segments give pain distributed over some part of the front of the thigh, knee, shin and ankle, while muscles supplied by the 5th lumbar and 1st and 2nd sacral segments give pain distributed over the buttock, the back of the thigh and calf, and the foot. Some typical examples are shown in Fig. 8. The segmental distribution of these pains in the leg has not been demonstrated so clearly as in the arm, but no pains have been observed with distributions inconsistent with a segmental pattern.

Comment. From the foregoing observations it would seem justifiable to conclude that, when saline is injected into a muscle, pain is referred to regions corresponding to the spinal segments from which its motor innervation is derived. In the limbs, the pain is only felt in a part of the available segmental regions unless it is severe. Each segmental region appears to consist of two component parts, one in the back, the other in the front of the trunk or in the limbs, and these parts seem to correspond with the anterior and posterior primary divisions of the spinal nerves.

It may be said that the distribution of referred pain arising from muscle follows a spinal segmental pattern. It is to be remarked that the pain felt is deep and diffuse and that the pattern does not correspond with the sensory segmental pattern of the skin as demonstrated by Head (2) or by Foerster (1).

Spread of pain between anterior and posterior divisions and between segments.

Anterior divisions. Referred pain derived from the musculature innervated by the anterior divisions is distributed in three ways. Pain arising from the intercostals and abdominal obliques is felt equally in back and front and seems to be distributed over a single segmental region. Pain arising from the rectus abdominis is felt largely in front, and seems to be distributed over the anterior regions of several segments, while the slight pain felt in the back is confined to a single posterior segmental region. Pain arising from the limb muscles is confined to the limbs and is only distributed over part of the available anterior segmental pain regions unless the pain is severe.

Posterior divisions. The musculature innervated by the posterior divisions can be divided into two groups ; the long muscles represented by the sacrospinalis, and the short muscles represented by the multifidus, the intertransverse, and other small muscles of the back. Pain derived from the two groups differs in its distribution. The long muscles give pain

almost entirely in the back and distributed over a wide area which seems to cover the distribution of several posterior segmental regions. From the short muscles pain is felt less extensively in the back but more in front, or in the limbs where it seems to be distributed over the anterior region corresponding to the segmental muscle stimulated. Fig. 9 illustrates results obtained from these two groups of muscle at the level of the 9th thoracic and 5th lumbar spines.
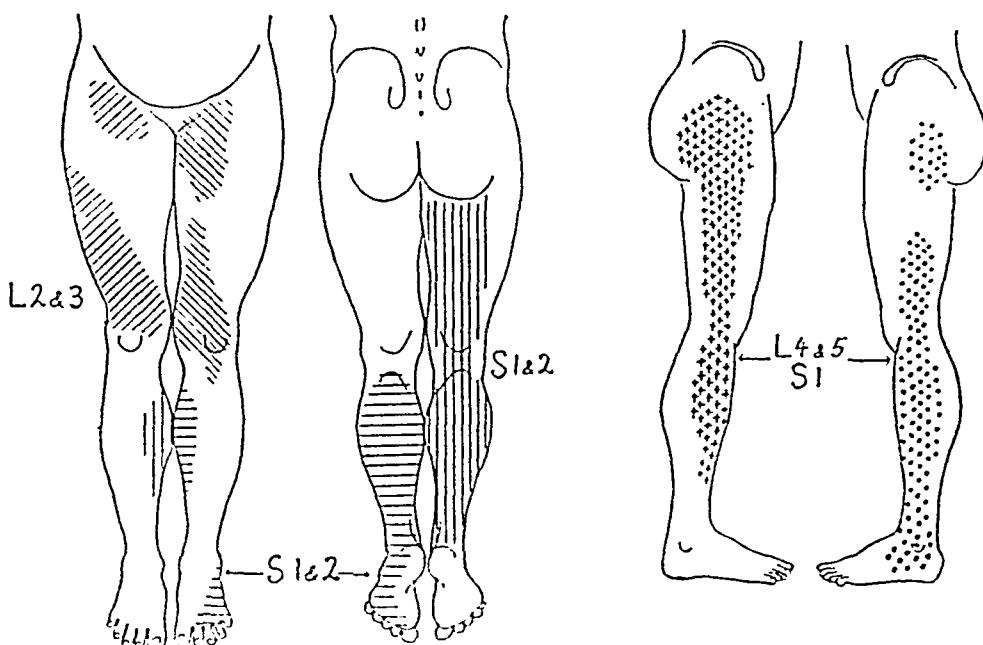

Fig. 8. Distribution of pain from leg muscles. From sartorius (right leg) and from adductor longus (left leg) oblique hatching; from gastrocnemius vertical hatching; from first interosseus horizontal hatching; from tensor fasciæ femoris crosses; from peroneus longus stippling.

The posterior segmental pain areas are found to correspond approximately with the distribution of the posterior primary divisions of the corresponding spinal nerves after they emerge from under cover of the muscles. This is shown in Fig. 10. The posterior divisions of the 6th, 7th, and 8th cervical nerves fail to emerge from under the muscle; and it is interesting to note that the pain produced by injecting 0·3 c.c. of 6% saline into the erector spinæ opposite the 7th cervical spine is felt in two regions, corresponding to the distribution of the 1st thoracic and to the 2nd, 3rd, 4th, and 5th cervical nerves, no pain being felt opposite the site of injection.

Intersegmental spread. The intercostal muscles probably represent the purest segmental musculature in the body; and the following observations were made to show the spread of pain arising from the intercostal spaces. Slight pains were produced from the 4th, 5th, and 6th intercostal spaces and their distribution in front and back noted. A very severe pain

was then produced from the 5th space and its distribution was found to cover the distribution of all three slight pains. This is shown in Fig. 4. The observation suggests that very severe pain derived from a muscle supplied by a single spinal nerve may spread to neighbouring segmental pain regions.
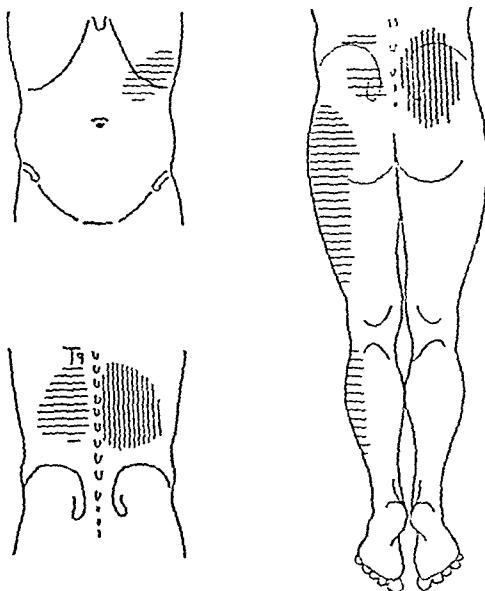

Fig. 9. Distribution of pain from *sacrospinalis* (vertical hatching) and multifidus (horizontal hatching) stimulated opposite the 9th thoracic and 5th lumbar spines.

Referred tenderness.

In this work on the distribution of pains arising from muscle it was often noticed that the region in which the pain is felt is tender to pressure ; this tenderness appears to lie deep to the skin.

The extensor digitorum communis in the forearm was chosen for further investigation. 0·2 c.c. of 6% saline when injected into the belly of this muscle in the forearm gives some pain in the forearm and severe pain over the back of the hand. The sensation on the back of the hand was tested before and after the injection of saline, and it was found that while the pain lasted there was slight tenderness to deep pressure and more definite tenderness to tapping but no skin tenderness could be detected. These observations were repeated using the brachialis anticus which gives pain in the region of the elbow and over the radial aspect of the forearm. The tenderness of the forearm has the same characteristics as those just described. The referred tenderness is found to correspond in distribution with the referred pain, except when this pain is felt deeply in the region of joints, and then the

tenderness is often absent. The tenderness also corresponds with the pain in duration and severity.

The idea that the tenderness is deep rather than cutaneous is confirmed by the following experiments. 1 c.c. of 2% novocaine is injected intradermally to give an analgesic area of skin about $2 \times 1\frac{1}{2}$ inches on the back of the hand. The tenderness resulting from saline injected into the extensor digitorum is undiminished beneath the area of analgesia skin. But if both skin and deep structures on the back of the hand are anæsthetised,
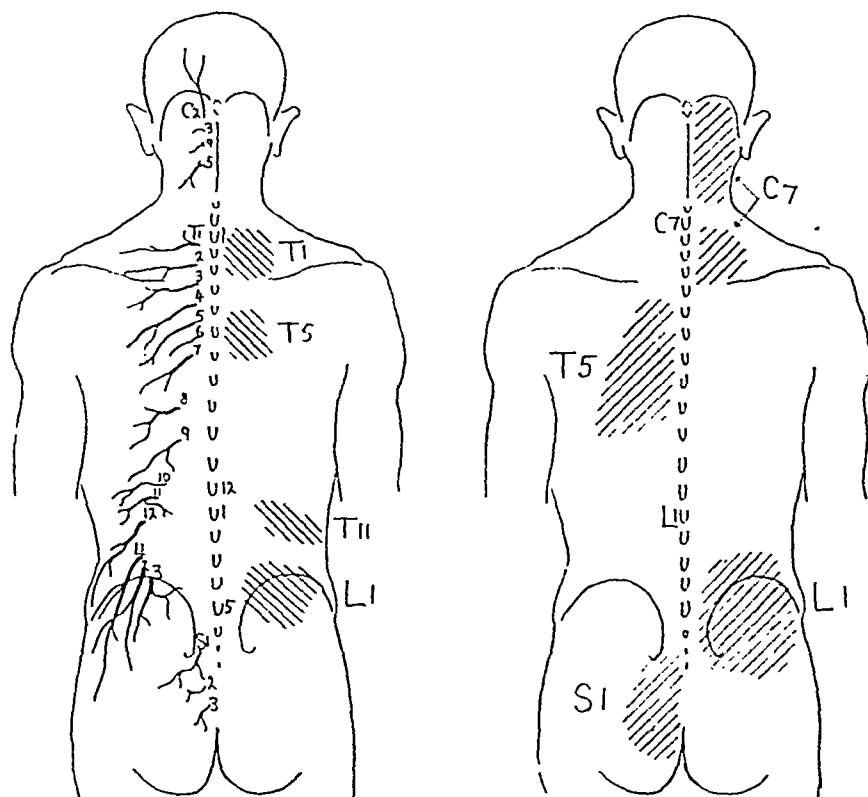
Fig. 10. Distribution of posterior pain areas and posterior divisions of spinal nerves. On left hand figure hatched areas represent pain referred from the front. On right hand figue hatched areas represent pain from sacrospinalis injected opposite the spine corresponding to the number noted.

the tenderness is no longer obtainable, though the severity of the spontaneous pain felt over the back of the hand remains undiminished. This is observed more clearly if similar pains referred to the anæsthetic and non-anæsthetic hands are compared simultaneously. This experiment is open to the criticism that the pain might be felt deeper in the hand than the anæsthetic had penetrated. This is avoided by using the flexor digitorum profundus which gives pain felt in the hand in the region of the hypothenar eminence and 5th knuckle. The ulnar nerve is blocked at the wrist with novocaine. When the anæsthesia and paralysis are full, 0·2 c.c. of 6% saline is injected

into the flexor digitorum profundus belly, and the resulting pain is felt clearly in the 5th knuckle and hypothenar eminence, although both these structures are completely anæsthetic.

From these observations it seems that referred pain from muscle is associated with tenderness of the deep structures rather than of the skin. If the region to which pain is referred is anæsthetised the tenderness is abolished but the pain remains unchanged. This suggests that the pain is independent of sensory impulses derived from the region of reference.

Comment. The results of these experiments may be discussed briefly in their relation to the mechanisms of referred pain.

The segmental distribution of muscle pain shows that it depends upon the architecture of the nervous system.

The observation that the pain is not abolished by anæsthetising the region of reference is inconsistent with the idea of an irritable focus in the cord, or with any reflex mechanism involving stimulation of the nerve endings in the area of reference, as under these theories the pain would be dependent upon sensory impulses derived from the area of reference.

What then may be the mechanism ? Two points seem to throw light on this problem. Firstly, the pain does not appear to be referred to areas of skin, but rather to deep structures such as the joints or testis ; and secondly pain arising from muscle appears to be confused by the subject with pain arising directly from these other structures. This suggests the possibility that the impulses responsible for pain from muscle and from the other deep structures, may follow a final common path in the central nervous system. That referred pain from muscle is always a diffuse pain might be explained by the pain fibres from muscle having diffuse instead of accurate synaptical connection with the central nervous system.

SUMMARY AND CONCLUSIONS.

1. An extensive investigation has been made of the characters and distribution of pain produced from accessible somatic muscles.

2. Fascia and tendon sheath give sharply localised pain, while muscle gives diffuse pain which is referred.

3. The diffuse pain from a given muscle is always distributed within certain regions, though the distribution within these limits varies from individual to individual, and according to the part of the muscle stimulated.

4. Pain from muscle may be confused with pain arising from other deep structures such as the joints and testis.

5. The distribution of the diffuse pain from muscle appears to follow a spinal segmental pattern. This pattern differs from that of the segmental innervation of the skin.

6. Referred pain from muscle is associated with referred tenderness of the deep structures.

7. The mechanism of reference is briefly discussed and it is suggested that the impulses responsible for pain from muscle and the other deep structures may follow a final common path in the central nervous system. The diffuseness of the pain might be explained by the.pain fibres from muscle having diffuse instead of accurate central synaptical connections.

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DELAY OF PAIN PERCEPTION IN TABES DORSALIS.*

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This paper deals with the delay which may occur in the perception of pain from a single stimulus in tabes dorsalis.

It was found by Lewis and Pochin (6) that a suitable stimulus to normal skin caused pain immediately, followed after a brief interval by a second pain. The delay of the second pain is due to its slow conduction in peripheral nerves, at a rate of about 1 metre per second.

This phenomenon of a double pain response to a single stimulus was noticed in 1884 by Rosenbach (14) although incorrectly explained by him. He examined the pain response from normal skin on account of observations by Remak (12) and others who had found a delay in pain perception in some cases of tabes dorsalis. Rosenbach identified this delayed pain in tabetics with his second pain from normal skin, but had no explanation for the failure of immediate pain or for the delay of the second pain. It seemed desirable to reinvestigate the delayed pain in tabes to determine if the amount of the delay is in fact equal to the delay of the second pain in normal subjects. It may be stated at once that this has been found to be the case.

The painful stimulus used was a prick with a needle mounted at right angles to a stiff wire. The prick was made through a sheet of tin foil spread on the surface of the skin, thereby completing an electric circuit and recording the instant of stimulation on a smoked drum. The subject, on feeling the delayed pain, pressed a morse key which also recorded on the drum.

The delay of the second pain in normal subjects was obtained in this way, but using a light prick with the needle. With heavy pricks the immediate pain becomes so strong that the delayed pain is often unrecognised. The results for the leg are given in Table I, each value being the mean of 3 to 5 records made after a few trial stimuli. It will be seen that the amount of the delay decreases as the stimulus is made higher up the limb, but that the values are only approximately equal in different subjects. As a sufficiently

exact measure of reaction time, the subject was given a tactile stimulus on the wrist, and instructed to press the key as soon as he felt the contact. These results appear in column 5 of the table.

TABLE I.

Delay in seconds.

Normal subjects (second pain).

Subject.	Site of stimulation			Reaction time.
	Toe.	Ankle.	Knee.	
1.	2·0	1·8	1·4	0·4
2.	2·0	1·8	1·2	0·3
3.	1·5	1·4	1·2	0·2
4.	1·7	1·0	0·7	0·3
5.	1·5	1·4	1·2	0·3
6.	1·9	1·7	1·3	0·4
Mean	1·8	1·5	1·2	0·3

Tabetics (pain).

Case.	Site of stimulation			Reaction time.
	Toe.	Ankle.	Knee.	
1.	1·8	1·5	1·0	0·4
2.	—	1·7	—	0·4
3.	1·5	1·2	1·1	0·4
4.	1·8	1·6	1·2	0·4
5.	1·8	1·8	1·1	0·8
6.	1·9	1·5	1·3	0·6
7.	1·6	1·1	—	0·6
8.	1·8	1·9	1·1	0·2
Mean	1·7	1·5	1·1	0·5

The delay of pain in tabetics was measured similarly, except that the needle prick was stronger. This was so because, from normal skin, a light prick may stimulate second pain endings, but be below the threshold for first pain and thus give a delay of pain perception. It is therefore necessary

to use in the tabetic a strong stimulus which would certainly excite the first pain endings in a normal subject, for otherwise a delay would necessarily be found equal to that of the second pain. As so used, the needle gave pricks with a thrust of 20 g., while in the normal subject the first pain is clear with pricks of a 2 gram thrust.

Twelve consecutive tabetics were investigated, in whom the diagnosis was clear, and of these seven experienced only a delayed pain in the legs. In these a needle prick was felt as a touch unaccompanied by pain, but followed after 1 to 2 seconds by a single burning pain which developed over a brief space of time and faded away, leaving a slight itch which could be abolished by rubbing. These are also the features of the normal second pain. The pain was apparently quite distinct, and responses in the absence
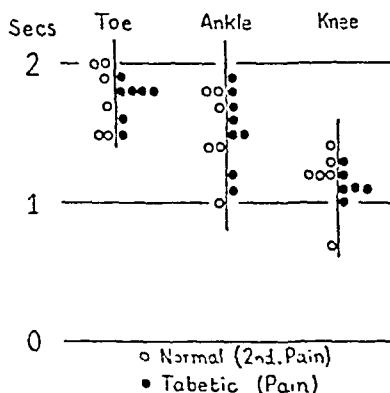
Fig. 1. Delay in perception of pain for different sites of stimulation, in normals and tabetics.

of a stimulus were hardly ever obtained. The pain, however, was described as dull if compared with immediate pain from unaffected skin. One further case of the twelve felt a needle as giving a slight prick when it touched the skin, followed by a burning pain after the same delay as for pain in the former patients. This case was not included in the series. The remaining four cases felt pain immediately. No case had complete pain loss from the legs except in one instance from the toes. In addition, one case was sent for and examined because she was known to have delayed pain perception. The results obtained from the eight tabetics showing the phenomenon of delay are given in Table I, Part 2, and compared in Fig. 1 with the delays from normal subjects. It will be seen that, for any one given region of the leg, the delay is about the same in different tabetics, and equal to the delay for second pain in normal subjects. Further, the delay decreases as stimulation is made higher up the leg, as is the case in normals.

It is obvious that the two series are in conclusive agreement. A minute comparison of differences is impossible for two reasons. Firstly, the routine stimulus was strong in tabetics, having a thrust of about 20 g., but weak

in the normal subjects, being varied to give a clear second pain sensation, and having a thrust of about 1 to 3 g.. Since the second pain develops to its full intensity over an appreciable time interval, the stage in this development at which it is recognised might depend upon its intensity. However, on comparing a series of light pricks and one of heavy pricks in a tabetic, the delay was found to be about equal for each.*

Secondly, the simple reaction times for tactile stimuli are on the average longer for the tabetics than for the normals. If this implies a difference in the interval between experiencing *pain* and pressing the key, then these reaction times should be deducted from the recorded delays before the comparison between normals and tabetics is made. Fig. 2 indicates this new comparison, and suggests that the small average difference is of doubtful
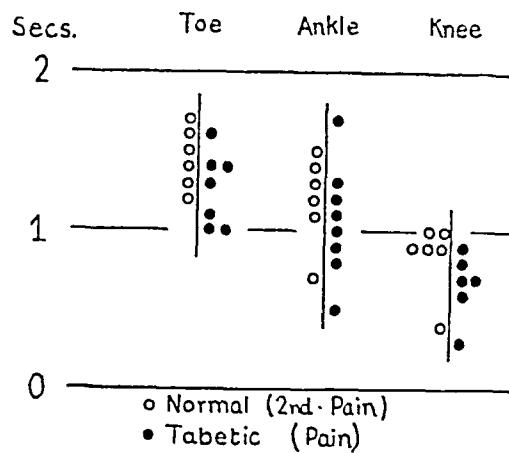
Fig. 2. As for Fig. 1 but subtracting reaction times from the recorded delays.

significance on the number of cases examined, as is indeed confirmed by calculation.

It can be stated therefore, that the delay in pain perception in tabes dorsalis is due to a slow peripheral conduction of the pain impulses at a rate corresponding to that of the known slowly conducting pain fibres from normal skin. And it is concluded that the appearance of the delay is due simply to a failure of the rapidly conducting group of pain fibres or their connections giving prominence to the effects of the slowly conducting group; and not to any region of pathologically slowed conduction.

The frequency with which the first pain sense is lost from the legs proved to be unexpectedly high; but unless sought for specifically the phenomenon is readily missed, since each prick produces a clear pain and no defect may be apparent or pain may simply be described as dull. In several of these cases the delay had not been noted in routine neurological examination.

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* This observation would not, of course, exclude the possibility that, even using equal stimuli, the second pain *sensations* might be unequal in the tabetic and the normal, which might produce a slight discrepancy in reaction times.

It is usually stated that the delay is revealed by stimulation of the legs. The question arises whether the loss of first pain sense is limited to the legs or is more generalised. Tests on the wrists and fingers revealed a loss of first pain sense in these positions in half of the cases showing the phenomenon in the legs. In such instances the delay was about 1 sec. from the wrist, and a little more from the fingers, corresponding to the delay of normal second pain. In one case the loss of first pain sense was confined to the ulnar border of the forearm and wrist.

Investigation of first pain loss from areas on the trunk is less simple owing to the small differences between the reaction time for first pain, and that for the normal second pain which on the trunk is one second or less. In no instance have greater delays been obtained from hypoalgesic areas on the trunk than are normal for second pain in such positions. It would be convenient if a stimulus could be found which would consistently excite the first pain system but not the second, so that the areas of first pain loss could be directly mapped. No such stimulus has been found although it seems likely that certain conditions of electrical stimulation might achieve this.

It may be noted that, after a little practise, it is simple to time delays approximately, using a watch ticking in fifths of a second. The ticks may be counted in fours, the prick being made on a "four", and the counting stopped when the patient is heard to say "Now!" The method is rapid, and times so obtained agree remarkably with the more reliable method used for these observations.

The sensory defects of the cases have been considered but it is impossible to make any general statement on the loss of first pain sense in relation to the order in which other sensations are lost.

It should be emphasised that the stimulus has been a single strong prick with a needle, first felt as pain after a delay, as observed by Remak (12, 13); to be distinguished from a phenomenon described by Naunyn (9, 10 and 11) that in tabes, repeated stimulation may begin to cause pain after a few seconds, although a single stimulus of the same strength is painless. Confusion has arisen because each author partly described also the phenomenon observed by the other. It should be clear too that this paper nowhere deals with prolongation of pain, but with delay in its onset. In previous case reports and in text books the amount of the delay is variously stated, but usually without evidence as to method of timing, stimulus or site of stimulation. The original systematic investigations include delays for other sensations particularly for temperature ; the average values found for pain are 1½ sec. from the leg by Leyden (7), 2½ sec. from the foot by Fischer (2) and also by Takács who observed this delay in 3 out of 8 cases of "ataxie," none of whom had delay from the arm.

While the phenomenon is usually observed in association with tabes dorsalis or other diseases of the spinal cord, Erb (1), Weir Mitchell (8), or in peripheral neuritis, it may also be found with local lesions of the peripheral

nerves. It is not easy to accept Kraussold's reports (3, 4, 5) of the development a few days after nerve suture of a long delayed pain with normal pain sense within one or two weeks ; but Erb (1) and Westphal (16) quote cases having local lesions in the arm, and pain delayed by 1 to 2 sec. and 1 to $1\frac{1}{2}$ sec., on pricking the skin of the hand.

It appears that, while there may yet prove to be instances in which delay of pain sensation is due to other factors, the usual cause is a failure of the rapidly conducting pain fibres, without a corresponding defect in the slowly conducting group.

SUMMARY.

The delay of pain perception in tabes dorsalis is due, not to an abnormal slowing of impulses in some diseased region, but to a defect in one group of pain fibres which conduct rapidly, revealing the effects of a second group which conduct slowly.

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UNILATERAL RETRACTION OF THE UPPER LID IN GRAVES' DISEASE.*

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OF the many eye signs described in Graves' disease, the two which are outstanding in clinical importance are the most frequently confused with each other ; firstly, exophthalmos, consisting of a protrusion forward of the eye from its normal position in the head ; secondly, Dalrymple's sign of retraction of the upper eyelid upward and backward over the eyeball, producing a characteristically staring eye with exposure of a band of sclera above the cornea, as seen in the following cases (Fig. 9). It is unfortunate that Dalrymple's sign creates an illusion of exophthalmos, especially when the eyes are viewed only from in front, as has been pointed out frequently. It will be shown that the two phenomena, exophthalmos and upper lid retraction, may appear separately in Graves' disease. Cases occur in which there is conspicuous exophthalmos, yet the upper lids are in a normal position relative to the cornea ; and other cases are seen in which the upper lid is retracted yet there is no exophthalmos, as measured instrumentally. In these observations, exophthalmos has been estimated by two devices, one a modification of Hertel's instrument, measuring the position of the corneal apex relative to the inferior orbital margin ; the other measuring more accurately, and from the lateral orbital margins. The words exophthalmos and proptosis are used indifferently to refer to a forward displacement of the eye.

Retraction of the upper lid, referred to subsequently as lid retraction, is described first. While frequent bilaterally in cases of Graves' disease, unilateral lid retraction is not commonly seen, but is of particular interest ; it allows the eyes and lids on the normal and abnormal sides to be compared in the same subject, and so may reveal phenomena associated with the retraction which are not obvious in the bilateral cases. The appearance of the eyes is described in detail in four cases of unilateral lid retraction in

The lower lid is a little raised relative to the cornea when the eyes are directed forward and upward and, with a position of the eyes in which the normal lower lid margin lies on the limbus, the opposite lower lid covers ½ to 1 mm. of cornea. It is difficult to be certain whether this is due to an elevation of the lower lid or to a downward displacement of the globe, or whether, as seems probable, both factors enter.

When the eyes are lightly closed, the free border of the affected upper lid remains higher than its fellow. The lower lid is usually brought up to meet it, and the palpebral fissure is thus higher on the affected side and straighter.

The upper lid in sleep remains retracted as in light closure. The lower lid, however, is not raised to meet it, so that a gap results (verified only for Cases 1 and 2). In full anæsthesia the appearance is the same (verified in Case 1 only).

When the eyes are closed, a conspicuous single fold of skin is present on the upper lid of the affected side. This fold is at a level of 6 to 10 mm. from the free border of the lid. Similar but bilateral wrinkling of the skin of the upper lids may be observed in some normal people so that its presence does not justify a diagnosis of lid retraction. In the present group of cases, about half showed some bilateral wrinkling. In all cases save one (*Case 6*) however, the wrinkling was obviously greater on the affected side. In one instance (Case 2) a wrinkle appeared when lid retraction developed on the same side (Figs 7 to 10).

In all the cases the appearance of the eye has strongly suggested unilateral exophthalmos, but in only one case has there actually been more than 1 mm. difference between the eyes. Such a difference is often found between the eyes of normal subjects by the same methods of measurement, but a slight prominence on the affected side has consistently been noted.

As the upturned eyes were rotated downward, the affected upper lid descended so as to expose sclera above the cornea during most of its descent.

It is evident that in Graves' disease lid retraction may occur unilaterally, or bilaterally as in Case 2, associated with only trivial exophthalmos. It is now to be considered whether exophthalmos can occur without lid retraction and if so, what is the appearance characteristic of this pure exophthalmos.

It is of interest to consider first the appearance of the eyes in a simple mechanical exophthalmos, although it will not necessarily be the same as in the exophthalmos of Graves' disease. When the position of the lids on the two sides is compared in a patient with unilateral exophthalmos of this kind, it is found that the upper lid maintains its normal relation to the pupil, even though the eye is proptosed by 10 mm. or more, the lid itself lengthening by several millimetres. The lower lid, however, does not lengthen in this way so that, withdrawn downward and under the proptosed eye, it reveals a wide band of sclera below the cornea. This appearance may

be obscured if the lids are œdematous, or if the optical axes are not parallel, but otherwise appears to hold true. Two cases seen recently illustrate this (Figs 17 and 18).

Case 7, a young man, developed a secondary deposit from a parotid tumour, producing exophthalmos of the left eye of 8 mm. as compared with the right (Fig. 17). There was some limitation of movement of the proptosed eye, which was displaced downward, but there was no œdema of the lids. Relative to the cornea, the left upper lid was normal in position, while the lower lid was 3 mm. lower on the eye than its fellow.

Case 8, a man of 34, had exophthalmos of the left eye of 12 mm. owing to pressure from an ivory osteoma arising in the ethmoid bone (Fig. 18). He had some limitation of movement of the left eye, which was displaced upward, but there was no œdema of the lids. This eye could be closed voluntarily but was not closed in sleep. The free border of the left upper lid stood at the same level on the cornea as that of the right, while the lid itself was 5 mm. longer than its fellow in sagittal extent. The left lower lid rested on the eye at a position 6 mm. below and behind that corresponding to the right lower lid.

Seale (3) quotes a case in which an ethmoid mucocoele had displaced the left eye 13 mm. forward, 10 mm. outward, and 5 mm. downward, yet a photograph shows the position of the left upper lid relative to the pupil to be normal, the lower lid exposing a wide band of sclera. The same disposition of the two lids is amply illustrated in a series of cases of unilateral exophthalmos described by Elsberg, Hare and Dyke (1).

Turning now to the exophthalmos of Graves' disease, the situation is less simple since in some cases it is to be expected that lid retraction may also be present. It is found, however, that cases are not uncommon which present an appearance exactly similar to that described for exophthalmos of mechanical origin ; such cases having conspicuous exophthalmos, yet with an upper lid in a normal position relative to the pupil. The lower lid is low, exposing a wide band of sclera below the cornea. The cases must presumably be regarded as having exophthalmos but without any retraction of the upper lid ; and also indicate that the actual protrusion of the eyes in Graves' disease probably gives rise to the same positions of the lids as it does when it arises from mechanical causes. Two such cases of exophthalmos without lid retraction may be described.

Case 9, a man of 21, had noticed palpitations, tiredness, shortness of breath and prominence of the eyes for about 7 years. On examination he was nervous and sweating, and had a flushed face. The thyroid was uniformly enlarged and the hands trembled when outstretched. The heart was enlarged, with a regular rate of 90 to 110 after a week in bed.

He had considerable exophthalmos, both eyes being advanced by 8 mm. from the approximate normal position. When the eyes looked forward the upper lids covered 2 mm. of cornea, while the lower lids exposed 3 mm. of sclera (Fig. 5). On his attempting to close his eyes, the lids did not quite meet, and the upper lids were unwrinkled.

After thyroidectomy the pulse fell to between 60 and 70. A section of the gland showed areas of moderate hyperplasia, with intervening colloid-filled vesicles and lymphoid aggregations.

Case 10, a man of 19, had developed a dislike for hot weather, with nervousness, palpitations and sweating for a year, during which time his eyes had "swollen." On examination he was flushed and sweating, and had a soft uniform enlargement of his thyroid. His heart rate was regular, and lay between 110 and 120 after several weeks in bed, the sleeping pulse rate being 10 beats lower.

The right eye was proptosed by about 5 mm., the left by 7 mm. from an average normal position. When the eyes were looking forward each upper lid covered 2 mm. of cornea, while the right and left lower lids exposed 2½ and 3½ mm. of sclera respectively (Fig. 6). The eyelids were unwrinkled when the eyes were closed, and the eyes were not closed in sleep. A month after thyroidectomy the pulse rate ranged between 75 and 90. A section of the gland revealed a considerable degree of epithelial hyperplasia.

Since it appears that the upper lid is not raised by exophthalmos itself in cases of mechanical origin and in some instances in Graves' disease, it is likely that in the remaining cases of this condition, in which it is raised, lid retraction is also present and accounts for the elevation. If this is so, the simple position is reached that lid retraction and exophthalmos may each be recognised in the presence of the other ; lid retraction by the height of the upper lid, and exophthalmos by the position of the corneal apex ; for exophthalmos does not cause elevation of the upper lid, and lid retraction only gives rise to slight exophthalmos.

The illusion of exophthalmos is apparently produced by the widening of the palpebral fissures which occurs in both conditions, whether it is due to an actual protrusion of the eyeballs forward between the lids, or to a retraction of the upper lids over the globe. But, while lid retraction gives rise to an illusion of exophthalmos, it does not cause the appearance which results when the eyes are in fact proptosed. In lid retraction the palpebral fissures are widened upward, sclera being visible above the cornea ; in exophthalmos they are widened downward, the low lower lids exposing an abnormal amount of sclera below the cornea. In the former case the abnormality is most evident when the patient looks downward ; in the latter case, when he looks upward. The closed eyes no longer appear prominent in lid retraction, and the skin of the upper lid is usually wrinkled. Eyes that are proptosed alone appear prominent when they are shut, and when they are viewed from the side or above ; and when the position of the corneal apex is measured relative to the bony wall of the orbit, it is found to be abnormal.

Since observation of cases of Graves' disease seems to indicate that lid retraction and exophthalmos occur and may vary independently of each other, a clear discrimination between the two is important if their respective clinical meaning or pathological significance is to be investigated.

SUMMARY.

1. Cases of unilateral retraction of the upper lid in Graves' disease show also on the affected side some elevation of the lower lid relative to the cornea, with a wrinkle of the skin of the upper lid when the eyes are closed (Figs 2 and 3).

2. They present an illusory appearance of unilateral exophthalmos (Figs 4, 7 and 11).

3. Cases of upper lid retraction show exposure of sclera above the cornea (Figs 9 and 13), while cases of exophthalmos show exposure of sclera below the cornea (Figs 5 and 6).

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Fig. 1.

Fig. 2.

Case 1.   Left lid retraction.

Fig. 3

Fig. 4.

Case 1.   (Cont.).

Fig. 5.
Case 9.
Exophthalmos.

Fig. 6.
Case 10.
Exophthalmos.

Fig. 8.
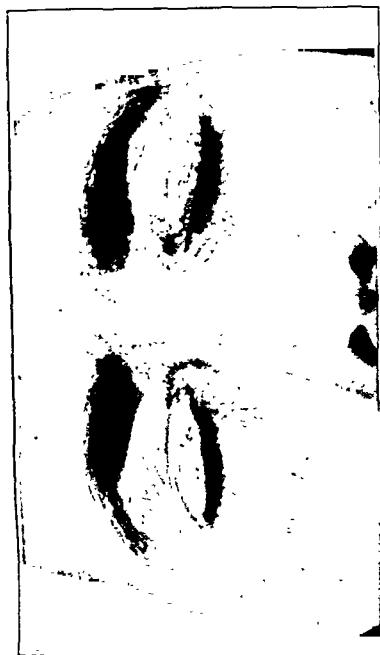
Case 2. January, 1937. Right lid retraction.
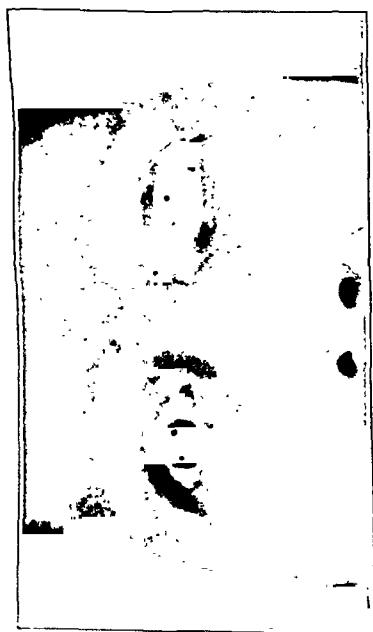
Fig. 10.

*Case* 2. October, 1937. Bilateral lid retraction.

Fig. 7.

Fig. 9.
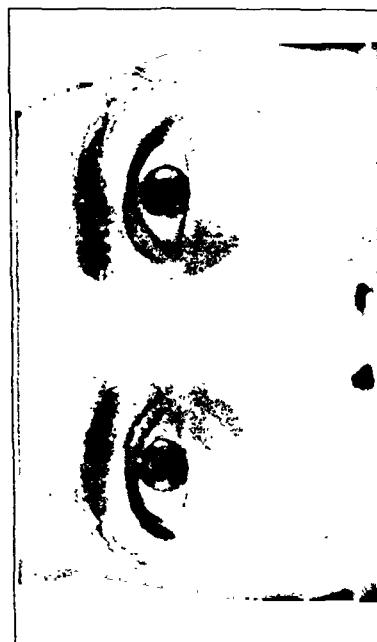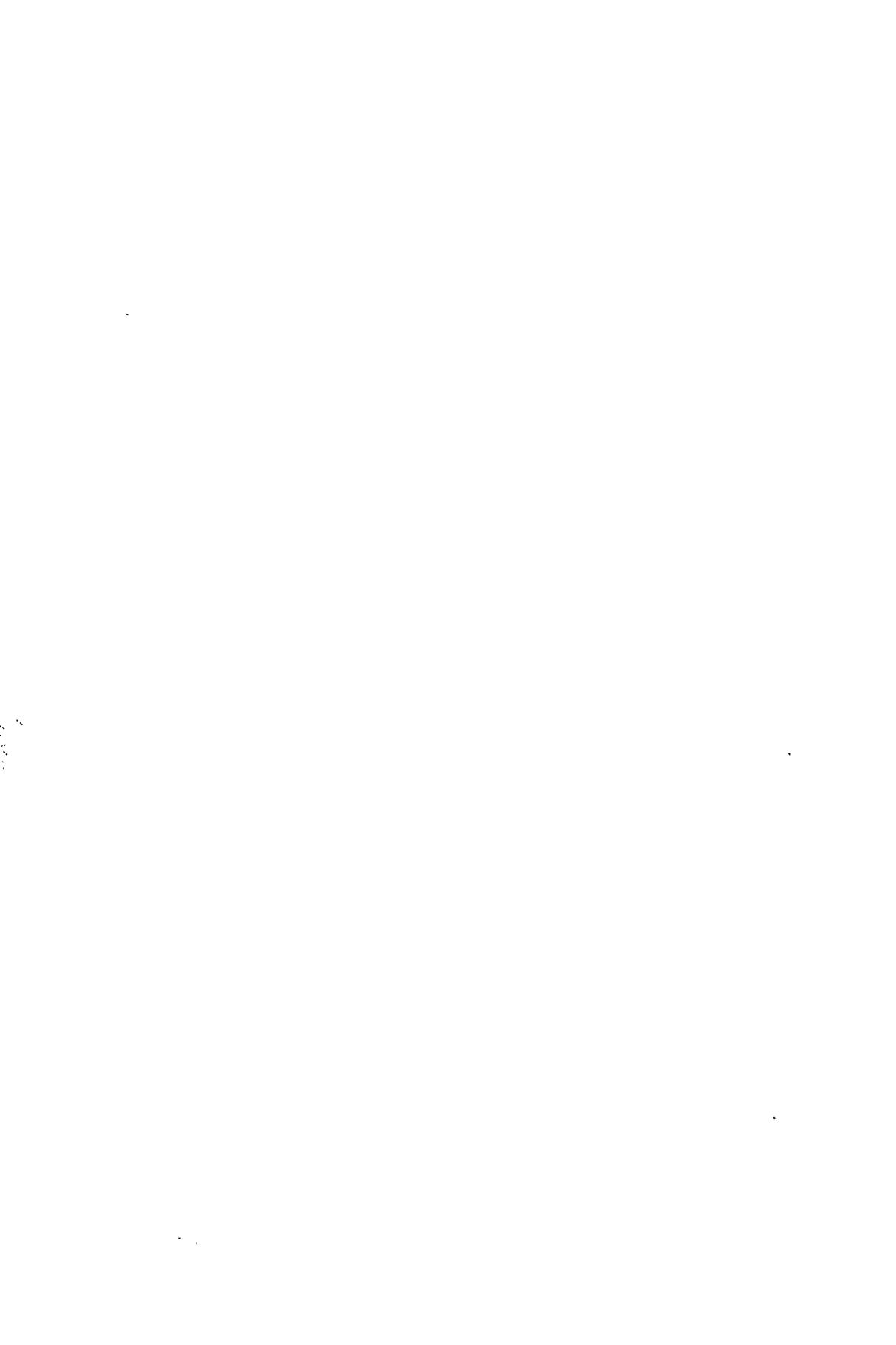
Fig. 12.

Fig. 11.

Case 4. Right lid retraction.

Fig. 11.

Fig. 13.

Case 5. Left lid retraction.

Fig. 16.

Case 6. Right lid retraction.
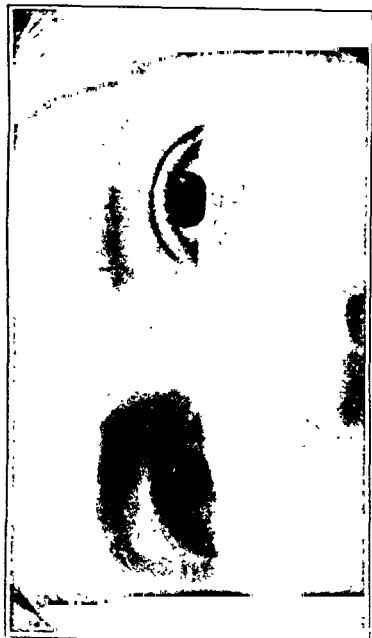
Fig. 18.

*Case* 8. Left mechanical exophthalmos.

Fig. 15.

Case 7. Left mechanical exophthalmos.

Fig. 17.

SOME OBSERVATIONS ON RENIN, A PRESSOR SUBSTANCE CONTAINED IN NORMAL KIDNEY, TOGETHER WITH A METHOD FOR ITS BIOLOGICAL ASSAY.*

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IN recent years experimental work has suggested that a chemical and not a nervous agent is probably responsible for the high blood pressure produced in the dog by constricting its renal arteries (3, 5, 6, 12), and possibly also for most forms of persistent high blood pressure in man (14, 15). In the experimental hypertension, and in chronic nephritis of man there are good reasons to select the kidney as the place to search for the supposed chemical agent.

A renal pressor substance was first described by Tigerstedt and Bergman (18) in 1898 ; it was found in saline extracts of fresh rabbit's kidney or of the dry residue obtained after treating rabbit's kidney with alcohol. The active substance, which they named " renin," was obtained from cortex but not to any appreciable extent from medulla of the kidney ; it was non-dialysable, stable at 56° but destroyed by boiling ; it was soluble in water, glycerine and in dilute salt solutions, but insoluble in acetone and in 50% and absolute alcohol. Injected into anæsthetised rabbits,‡ active extracts gave a rise of blood pressure beginning in 10 sec., maximal after 2 min. and lasting as long as 20 min. ; the rise was not abolished by section of the cervical spinal cord, nor by destruction of the spinal cord. Renin had no effect on the isolated heart. Tigerstedt and Bergman's findings were confirmed by Bingel and Strauss (1) who chiefly used renal press juice, and who stated that renin was destroyed by alcohol, acetone, acids and alkalies and was precipitated by 7/12 saturation with ammonium sulphate. Bingel and Strauss's purest preparation was obtained from autolysed renal press juice by fractional precipitation with ammonium sulphate. Other workers

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to obtain pressor effects in the anæsthetised animal from saline extracts of kidney were Vincent and Sheen (19), who obtained small pressor responses as well as depressor responses from saline extracts of many tissues, and Shaw (17).   On the other hand Pearce (13), using saline extracts of rabbit, dog and cat's kidney, and Hartwich and Hessel (10) using press juice of pig kidney were unable to obtain significant pressor responses in the anæsthetised animal.   Hartwich and Hessel (11) obtained pressor responses from renal press juice autolysed under xylene for 14 days, but the pressor substance or substances were probably amines produced by decomposition and had not the properties attributed to renin by Tigerstedt and Bergman and by Bingel and Strauss.

An attempt to find alterations in the renin content of tissues in renal disease was made by Bingel and Claus (2) who failed to find any renin in the kidneys of rabbits poisoned by corrosive sublimate or in the blood of " nephritics."   Later Hartwich (9) claimed to have produced hypertension in dogs by ligaturing one renal artery, and observed that press juice obtained from an ischæmic kidney 4 days after ligature of its artery and vein was pressor ; but he was unable to say whether this effect was greater than that of a similar preparation from normal kidney.   Prinzmetal and Friedman (16) have made more convincing observations, finding that in dogs whose blood pressure was raised by constricting one renal artery, saline extracts of the ischæmic kidney gave greater rises of blood pressure when injected into unanæsthetised dogs than did saline extracts of the unoperated kidney ; they also observed that saline extracts of human kidneys obtained at autopsy had, in general, a greater pressor effect in the unanæsthetised dog when the patients had had raised blood pressure during life than when the blood pressure had been normal.   Harrison, Blalock and Mason (8) observed that when dogs were rendered hypertensive by constricting one or both renal arteries, or by ligaturing the ureters, saline extracts of the affected kidneys produced greater rises of blood pressure than did saline extracts of normal dog's kidneys.

Unfortunately, during the last 20 years, there have been many claims as to the abnormal presence of pressor substances in the tissues of hypertensive patients and animals, which have not been substantiated by further work.   And although there are unusually strong indications that a renal pressor substance may be the agent responsible for certain forms of hypertension, there are several obstacles in the path of those who wish to test this hypothesis.   Thus, the failure of many workers to find any evidence of a pressor substance in normal kidneys has raised considerable doubt as to the existence of renin, and has led to the view, to some extent fostered by the work of Hartwich and Hessel (11), that the pressor substances found by some are the products of tissue autolysis or of bacterial action.   Again, the kidneys, like most other organs, contain abundant depressor material, and it is necessary to find some method of getting rid of this before the supposed pressor substance can be satisfactorily investigated.   Lastly, there is as yet

no method of assaying " renin." The importance of having a method of assay needs no emphasis, for it is largely to such methods that we may attribute the growth of knowledge concerning the rôle played by biologically active substances in health and disease.

The present work is an attempt to overcome these difficulties.

Methods of testing and preparing renal extracts.

Our earliest preparations were tested on the arterial pressure of cats anæsthetised with urethane and were invariably depressor. Later we obtained pressor responses with similar preparations on the unanæsthetised rabbit, and we have since used this method of testing renal extracts as a
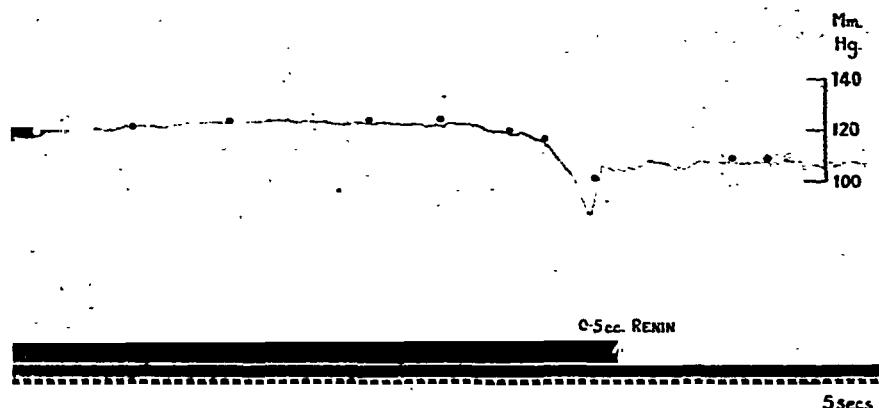
Fig. 1. Reads from right to left. A manometric blood pressure record from the femoral artery of a rabbit anæsthetised with sodium luminal (0·25 g. per kg.). The white dots superimposed on the record represent readings of systolic pressure in the central artery of the ear obtained by the capsule method at corresponding times. The arrow marks an injection of 0·5 c.c. of Ringer extract of the alcoholic residue from rabbit's kidney (10 c.c. Ringer per g. residue).

routine. The injections are made intravenously into the denervated ear of a warm unanæsthetised rabbit, the systolic pressure being measured on the other ear at intervals of 20 to 30 sec. with the capsule described by Grant and Rothschild (7). Provided that precautions are taken to keep the ear flushed, estimations of systolic pressure obtained from the central artery of the ear are, as Grant and Rothschild pointed out, in close agreement with the mean arterial pressure recorded by direct cannulation of the femoral artery (Fig. 1).

Tested on the unanæsthetised rabbit, saline extracts of rabbit's kidney, were chiefly depressor in doses corresponding to less than 0·25 g. kidney, and in larger doses produced death in about 4 minutes, preceded by

blanching of the ears and convulsions. We succeeded in obtaining extracts with a pure pressor effect by two methods. The first method, based on one described by Tigerstedt and Bergman (18), consists in treating the kidney with alcohol, and subsequently extracting the dry residue with Ringer solution. The second method is a preparation of the total globulin fraction of the kidney proteins. The methods as finally adopted for quantitative work are as follows :—

(1) *Alcohol method.* Fresh rabbit's kidney is pulped in a mortar and weighed into a boiling tube. 2 c.c. absolute alcohol per g. kidney are added, the tube is shaken to mix its contents thoroughly and put into the refrigerator for 24 hours. The mixture is filtered, small quantities of the residue adhering to the tube being detached with a stirring rod and washed with some of the filtrate into the filter paper. When all the residue has been transferred to the paper and no more alcohol is filtering off, the paper is wrapped in muslin and squeezed dry. The filtrate is discarded. The residue is transferred to a large filter paper on which it is spread and allowed to dry in a warm room (20 to 24°C.). The dry residue is powdered in a mortar and weighed ; about 0·6 g. is weighed into a boiling tube to which 10 c.c. Ringer solution per g. of powder are added. The mixture is agitated gently once or twice and the tube placed in the refrigerator for 24 hours. It is then removed and gently agitated several times during 10 minutes to mix its contents, which are then filtered through glass wool and muslin, the residue caught in the latter being squeezed dry. The solution, which is yellow and slightly turbid, is ready for testing.

(2) *Total globulin method.* Fresh rabbit's kidney is frozen solid with CO_2 snow, and after $\frac{1}{2}$ to 1 hour thawed out at room temperature. The kidney is finely pulped in a mortar and weighed in a boiling tube to which 4 c.c. Ringer solution per g. of kidney are then added. The tube is shaken for half an hour by a mechanical device and then allowed to stand for $1\frac{1}{2}$ hours in the refrigerator. The mixture is centrifuged and the supernatant liquid poured off into a graduated cylinder to which an equal volume of saturated ammonium sulphate is added. The mixture after standing overnight in the refrigerator is filtered and the precipitate is placed inside a cellophane* dialyser. After dialysis for 2 hours against running tap water and overnight in the refrigerator against tap water or Ringer's solution, the content of the dialyser is transferred to a graduated cylinder and is ready for testing. This preparation is a greyish opaque liquid.

Such extracts give a pure pressor effect in the unanæsthetised rabbit (Fig. 2). The rise of blood pressure is peculiar in being of slow development, beginning in about 10 to 15 seconds, and reaching its height in about 2 minutes. After rises of about 30 mm. Hg, the blood pressure usually returns gradually to normal in about 30 minutes ; with rises of 50 mm. Hg the response may last 45 to 60 minutes. At the height of the blood pressure

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* The cellophane used throughout this work has been number 300.
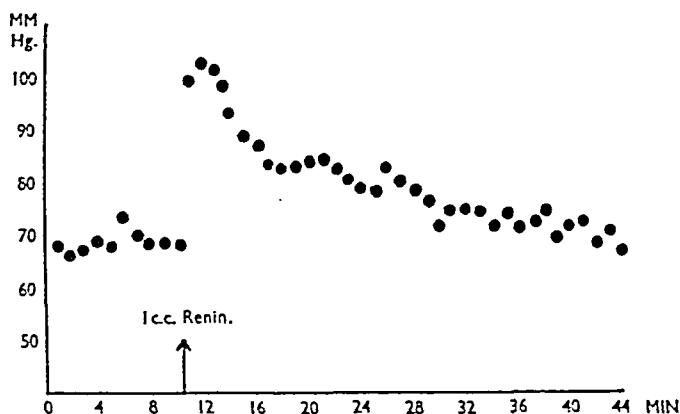
Fig. 2. A systolic blood pressure record obtained from an unanæsthetised rabbit by the capsule method. The arrow marks the intravenous injection of 1 c.c. of rabbit's kidney extract prepared by the alcohol method.

rise the pulse in the unanæsthetised animal is slowed. It is noteworthy that even when the blood pressure has been raised by 50 or 60 mm. Hg, there is little change in the appearance of the animal* which, if allowed free, goes about its ordinary occupations in an apparently normal manner. We have never seen any toxic effects from extracts made by the alcohol method, and rarely from the total globulin extracts.

A record obtained from a dog anæsthetised with nembutal is shown in Fig. 3.
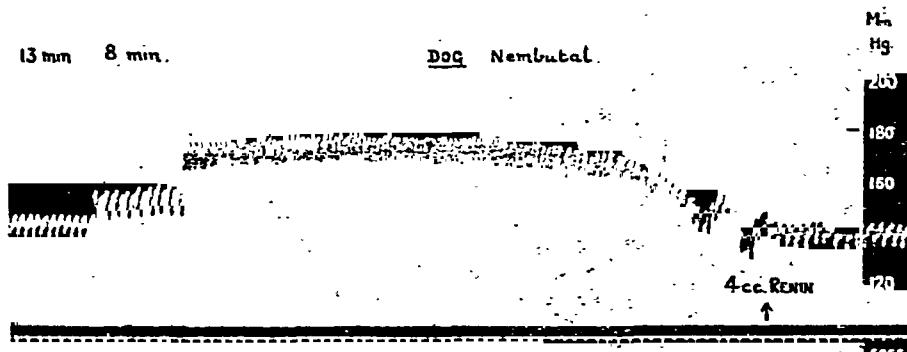
Fig. 3. Reads from right to left. A manometric blood pressure record from the carotid of a dog anæsthetised with nembutal. The arrow shows the injection of 4 c.c. of a purified preparation of renin from rabbit's kidney.

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* In this respect renin contrasts with tyramine and adrenaline, which produce conspicuous blanching of the rabbit's ears when doses giving comparable rises of blood pressure are used. With renin, a faint paling of the ground-tone of the ears is noticed when doses are used giving rises of blood pressure of 50 mm. Hg or less ; with larger doses the blanching of the ground-tone is definite.

Extracts prepared by one or both of these methods from the fresh kidneys of over 100 normal rabbits have invariably raised the unanæsthetised rabbit's blood pressure. Pressor responses have also been obtained in the rabbit from similar renal extracts made shortly after death from pig, dog, cat and man ; two kidneys from the cow and one from the horse gave inactive extracts. If cortex and medulla are carefully separated by dissection, and extracted separately, the cortical extracts are pressor, while the medullary extracts are inactive (Fig. 4). Extracts prepared by the total globulin method from brain, muscle, gut, suprarenal, spleen and blood of normal rabbits are inactive ; from normal rabbit's urine such extracts are usually depressor.
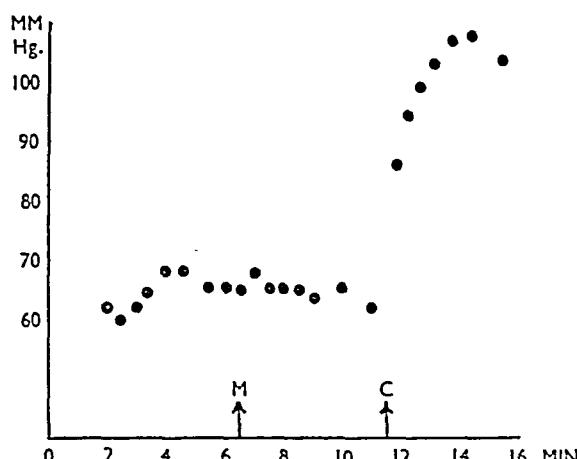

Fig. 4. A record of systolic arterial pressure from an unanæsthetised rabbit by the capsule method. At *M* 1 c.c. of an extract of renal medulla and at *C* 1 c.c. of an extract of renal cortex were injected intravenously. The extracts were prepared simultaneously from a pair of dog's kidneys by the alcohol method.

It is evident, then, that a substance raising the unanæsthetised rabbit's blood pressure is constantly present in the cortex of the kidney of many normal mammals. It is not present, at least in comparable quantities, in the other tissues examined. This substance, renin, is not a product of tissue autolysis or of bacterial action since it may be obtained from fresh kidney by the alcohol method which precludes such change.

Some chemical properties of renin.

In this work we have used chiefly fresh rabbit's kidney and extracts prepared in the ways already described. We have not specially investigated renin obtained from other species.

Stability. Provided it is kept cool, and in a suitable medium, renin seems to be relatively stable. Thus in the intact kidney kept in the carcase at about 10°C., the amount that can be extracted by the alcohol method is unchanged after 24 hours ; after 48 hours the kidney stinks and its renin content is diminished. The dry residue obtained after alcohol extraction

keeps without change for several days at room temperature, and for at least 12 months in sealed tubes in the refrigerator. In Ringer's solution the stability of renin depends largely on pH and bacterial action. Thus Ringer extracts of the alcoholic renal residue, made without aseptic precautions, maintain their activity for a week in the refrigerator; after 6 months the extracts stink and have lost most of their activity. At pH 4·0, the renin content is not detectably altered after 2 months in the refrigerator; but at pH 10 all activity disappears in 2 weeks.

Heat. Renin is thermo-labile. Boiling for 5 minutes or maintaining at 60°C. for 2 hours inactivates previously active solutions (pH 6·9) with the formation of a coagulum.

Solutions of renin can be concentrated without loss of potency by evaporation in vacuo at 25°C.

Acid and alkali. Ringer extracts of the dry alcoholic renal residue are faintly acid (pH 6·2 to 6·9), and are yellow and slightly turbid. Addition of acid or alkali to bring the pH to 5·0 or 9·0 produces a precipitate which · in the case of acid, contains a little, and in the case of alkali, none, of the original activity. If neutralised at once, the supernatant fluids, which are now clear, retain most of the activity of the original solution. As has been mentioned, renin is stable for long periods in weakly acid (pH 4 to 5) but not in alkaline (pH 10) solution. Addition of HCl or NaOH to a concentration of $\frac{N}{10}$ destroys all activity within 2 hours.

Dialysis. Renin does not pass through cellophane, and may be freed from crystalloids by dialysis.

Salting out. Bingel and Strauss (1) obtained their purest preparations from autolysed renal press juice from which they precipitated inactive material by 1/3 saturation with ammonium sulphate and all the active material by 7/12 saturation. Using fresh saline extracts we have found that the precipitate obtained by 1/3 saturation contains part, and that obtained by ½ saturation with ammonium sulphate, all of the activity of the original solution, provided the mixtures are allowed to stand for some hours in the refrigerator before filtering; the precipitates are redissolved and freed from ammonium sulphate by dialysis before testing. The total globulin fraction so obtained, after prolonged dialysis against distilled water and adjustment to pH 5·5, throws down a precipitate of euglobulin which may be centrifuged off and redissolved in Ringer's solution at pH 7. Most of the renin is found in the supernatant fluid (pseudo-globulin fraction) but some remains with the euglobulin.

Organic solvents. Renin is insoluble in alcohol, acetone, ether and chloroform. Alcohol and acetone at room temperature both tend to destroy the activity, but their action in this way is greater with some preparations than with others. On pulped kidney the inactivating action seems to be least pronounced. Thus, if kidney pulp is allowed to stand in the refrigerator with 2 volumes of absolute alcohol for 3 weeks, a good deal of renin may still

be recovered by extracting the dry residue with Ringer; the amount recovered is slightly less than if kidney and alcohol have only been in contact for 24 hours, and distinctly less than when the contact has been for a few hours only. When pulped kidney is treated with alcohol or acetone at room temperature and allowed to stand for 24 hours in the refrigerator, then the amount of activity recoverable from the dry residue is largest when 2 or 4 volumes of absolute, or 4 volumes of 80% alcohol are used, and slightly less when 2 volumes of acetone or 10 of absolute alcohol are employed.

From Ringer solutions of the dry alcoholic renal residue, precipitation by alcohol and subsequent extraction of the dry precipitate with Ringer give rather variable, and rarely quantitative recovery of renin. Inactive precipitates are always obtained with 30% and often with 50% alcohol; by raising the concentration of alcohol to 90%, renin is always precipitated from the 30% and usually from the 50% solution, but the recovery is usually small. It would appear then that renin is slightly soluble in 30% and possibly in 50% alcohol, but the inactivating action of the solvent makes an exact statement impossible. Maximum yields are obtained by precipitation with concentrations of alcohol of 80% or more.

With Ringer extracts of fresh kidney or its total globulin fraction, it is similarly impossible to obtain quantitative recovery of renin by alcohol precipitation at room temperature, particularly when the contact has been prolonged. Thus by extracting with Ringer the dry precipitate obtained from the addition of 6 volumes of alcohol to such a solution, less than 50% of the renin is recovered if the contact with alcohol has been made as short as possible, and less than 25% when the alcohol and renin have been in contact for 24 hours.

Lastly, if a chilled solution of renin is slowly added to 10 volumes of absolute alcohol at —10°C., the precipitate separated by centrifugalisation or filtration and washed with well cooled dry ether in a cold room (3°C.) and transferred to a vacuum desiccator, then renin may be recovered quantitatively from the dry precipitate by extraction with Ringer.

Adsorbents. Ringer solutions of the alcoholic renal residue may sometimes be decolourised without loss of potency by shaking with small quantities of charcoal (Norit). Larger amounts of Norit (over 1 g. per 100 c.c.) remove renin also; the adsorbtion of renin by charcoal is rather greater at pH 5 than at pH 8. Dialysed iron in small amounts removes some protein without the renin; in larger amounts the renin is also removed.

Discussion of chemical properties. The chemical properties of renin contrast sharply with those of all other known pressor substances with which renin cannot, therefore, be identical. Of known depressor substances kallikrein resembles renin chemically, but the two are not identical, for kallikrein (Padutin) lowers the blood pressure of the unanæsthetised rabbit.

The behaviour of renin to heat, semipermeable membranes, solvents and concentrated salt solutions strongly suggests that it is a protein. All our active preparations, however purified, have given a precipitate with trichloracetic acid, and, when their salt concentration was adequate, a coagulum on boiling.

The effect of anaesthesia on the action of renin.

It is surprising that renin has been found by so few workers, although its discovery dates only 4 years after that of adrenaline, and despite the obvious interest of a renal pressor substance both to pathology and physiology. This has not been due to misconception of its chemical properties, many of which were described by Tigerstedt and Bergman (18). It has been due, we think, to the customary use of anaesthetics in testing for vaso-active substances ; for the action of renin is reduced or abolished by these.

It has been remarked that our earliest renal extracts tested on the urethanised cat were depressor. For some time we were unable to account for our failure to demonstrate renin in this animal preparation.* Later, we were surprised to find pure depressor responses in a urethanised rabbit from renal extracts which we knew to be pressor in the unanaesthetised animal. Subsequent deliberate experiment has shown that urethane, ether, and nembutal reduce the pressor effects of renin in the rabbit. Illustrative experiments are as follows :

Protocol 1. Male black rabbit weighing 3·0 kg.. The left ear was anaesthetised with 2% novocaine injected into its base, and received the intravenous injections. The blood pressure was measured on the right ear by the capsule method. The responses to 0·5 c.c. of a Ringer extract of the dry alcoholic residue of rabbit's kidney, and to 4 mg. of tyramine acid phosphate dissolved in 1 c.c. of Ringer solution, are shown before and after anaesthetisation with urethane.

Time		Injection.	Initial B.P. mm.Hg.	Change in blood pressure mm.Hg.	
Hr.	Min.			Fall.	Rise.
0	34	0·5 c.c. Renin	88	0	43
1	18	4 mg. Tyramine	101	0	33
1	24	6 g. Urethane (subcutaneous and intraperitoneal injection of a 25% sol.).			
2	3	4 mg. Tyramine	82	0	22
2	8	0·5 c.c. Renin	70	14	30
2	40	4 mg. Tyramine	68	0	36
2	49	0·5 c.c Renin	60	13	25
3	51	4 mg. Tyramine	59	0	33
4	6	0·5 c.c. Renin	67	29	8
5	14	4 mg. Tyramine	50	0	25
5	37	0·5 c.c. Renin	52	21	0

* At one time we were inclined to believe that the cat would not respond to renin. We have since found that beautiful pressor responses may be obtained in the cat decapitated under ether and from which the anaesthetic has been removed by prolonged ventilation.

Protocol 2. Male brown rabbit weighing 2·5 kg.. The left ear was anæsthetised with novocaine, and received the intravenous injections, the arterial pressure being measured on the right ear. The renin solution was a preparation of the pseudoglobulin fraction obtained from rabbit's kidney. The amount of nembutal injected intravenously (2·3 c.c. in 3 hrs) was sufficient to keep the animal lightly anæsthetic, the corneal reflex being just obtained.

| Time | | Injection. | Initial B.P. mm.Hg. | Change in blood pressure mm.Hg. | |
Hr.	Min.			Fall.	Rise.
1	11	1·25 c.c. Renin	72	0	22
1	50	1·25 c.c. Renin	75	0	37
2	29	1·25 c.c. Renin	75	0	30
2	35	Anæsthetised with nembutal.			
3	7	1·25 c.c. Renin	76	0	21
4	25	1·25 c.c. Renin	71	0	13
5	38	1·25 c.c. Renin Allowed to recover from anæsthesia.	72	0	12
23	0	1·25 c.c. Renin	73	0	29

Protocol 3. Male brown rabbit weighing 2·5 kg.. The general procedure and renin solution used were the same as in Protocol 2, except that after the third injection of renin this animal was anæsthetised with ether. The corneal reflex was absent during anæsthesia.

| Time | | Injection. | Initial B.P. mm.Hg. | Change in blood pressure mm.Hg. | |
Hr.	Min.			Fall.	Rise.
0	14	1 c.c. Renin	73	0	25
1	18	1 c.c. Renin	72	0	26
2	6	1 c.c. Renin	78	0	27
2	12	Anæsthetised with ether			
2	53	1 c.c. Renin	77	0	14
4	40	1 c.c. Renin	76	0	16

Protocol 1 shows the progressive disappearance of the pressor and development of a depressor response to a renal extract after injecting urethane ; in this instance a Ringer extract of the alcoholic residue from rabbit's kidney was used. At a stage of anæsthesia when the pressor response to renin had almost disappeared the response to tyramine was not definitely altered. Protocols 2 and 3 show that nembutal and ether also reduce the pressor effect of renin. In these two experiments the extract used was the pseudoglobulin fraction obtained from a saline extract of kidney and no depressor effect was observed.

That anæsthetics so different chemically as ether, urethane and nembutal, interfere with the pressor action of renin is of theoretical as well as historical interest. The reason why they do so is outside the scope of this paper, but it is relevant to recall that Tigerstedt and Bergmann (18) and Bingel and Strauss (1) obtained pressor responses from renin in animals with their spinal cords destroyed.

Depressor effects of renal extracts.

In the urethanised animal, as we have seen, Ringer extracts of the alcoholic residue give a pronounced fall, which may or may not be followed by a rise, of blood pressure. This depressor response is due in part to a substance which is not renin, for it is stable to boiling for 5 min. and is removed by quantities of charcoal too small to affect the renin content, as judged by the rise of blood pressure produced in the unanæsthetised rabbit. The remaining depressor effect is due to a substance which behaves like renin in being removed by larger amounts of charcoal and in being destroyed by boiling, and which reacts like renin in respect to half saturation with ammonium sulphate and subsequent dialysis. Whether this substance is renin, or only something which closely resembles it in certain chemical respects, we cannot say. Although, as has been seen, the pseudoglobulin fraction of the kidney proteins produced pure pressor responses in rabbits anæsthetised with ether or nembutal, none of our extracts containing renin, however purified, have failed to give depressor responses in the animal anæsthetised with 2 g. urethane per kg.. Kallikrein may well be present in our preparations of renin since they behave rather similarly to adsorbents, heat, and dialysis, and are both insoluble in alcohol and concentrated salt solutions.

The assay of renin.

Comparison of the renin content of two solutions. Tigerstedt and Bergmann (18), and Bingel and Strauss (1) both stressed the absence of any relationship between the amount of a given renal extract injected and the resultant rise of blood pressure in anæsthetised animals. Anæsthetics, however, as we have shown, seriously interfere with the action of renin. In most unanæsthetised rabbits of quiet disposition, repeated injections of a given dose of renin will produce rises of blood pressure which do not differ from one another by more than a few millimetres of mercury, provided that the blood pressure is allowed to return to its resting level before each injection (for example, Protocol 3). In such rabbits also, the rise of blood pressure increases with increasing dosage of renin. The relationship between size of dose and size of response is not linear but is usually of a form shown by Fig. 5. When the dose is plotted on the logarithmic scale as in Fig. 6, the relationship is approximately linear. A similar relationship between response and the logarithm of the dose is found with most biologically active substances (4).

to estimate about 4 renin preparations on the same day, and experiments are usually arranged with this in view. Just before the solutions are ready to test, 5 or 6 rabbits, prepared as described, are put in a run in a warm room at about 24°C. in order to keep their ears flushed. The first rabbit to be used sits on a comfortably warm pad and is firmly covered with a blanket secured at the sides by sandbags. It should remain quiet in this position. The ears being fully flushed, the blood pressure is recorded by Grant and Rothschild's capsule at 20 to 30 sec. intervals from the right ear; when steady readings have been obtained for 3 minutes, an injection of one of the unknown renin solutions is made into a vein of the denervated left ear. The insertion of the needle should not affect the blood pressure. Records of blood pressure are continued until the rise is over, that is to say for about 3 minutes after the injection. The rabbit is then returned to the run and a second put on the warm pad, and the response of this rabbit to a second unknown renin preparation is recorded. The rabbits are used in rotation, so that by the time any rabbit's turn comes for a second injection its blood pressure has returned to normal. The responses to unknown and to standard preparations are determined alternately in each rabbit, until doses are found giving identical responses. These responses should lie between 20 and 40 mm. Hg. The extracts should be diluted so that the volume injected lies between 0·5 and 2·0 c.c.; within this range the response is not influenced by the volume of fluid introduced.

To assay 4 solutions in this way takes usually from 4 to 6 hours. It is advisable to have more rabbits than solutions to be assayed, as one or more rabbits are sometimes found to be unreliable on that day, giving irregular responses which do not increase with increasing dosage. Some rabbits prove persistently unreliable and are discarded. Reliable rabbits may be used repeatedly for injection until their veins become obliterated.[*] We have seen no evidence that rabbits develop either tolerance or increased sensitivity to renin injections (from rabbit's kidney) repeated over several months.

Examples of a satisfactory and of an unsatisfactory assay may be given.

1. March the 6th, 1937. Rabbit 53 was injected with the standard solution and with extracts made by the alcohol method from the kidneys of rabbits 147 and 139. The results were as follows :—

Dose injected. c.c.	Rise of blood pressure (mm. Hg) produced by :—		
	Sol. 147	Sol. 139	Standard sol.
0·15	—	—	17
0·25	21	19	28, 28
0·4	28	23	—
0·5	31	28	—

From these results we may judge that 0·25 c.c. of the standard solution contains as much renin as 0·4 c.c. of solution 147 and 0·5 c.c. of solution 139. Solution 147 thus contains 0·625 units per c.c., and solution 139 0·5 units per c.c..

2. *October the* 6th, 1937. Rabbit 123 was injected with two solutions; the results were :—

Dose injected. c.c.	Rise of blood pressure (mm. Hg).	
	Sol. H.	Standard sol.
0·25	23	26
0·37	25	25

The absence of any increase in response in spite of a 50% increase in dosage of the standard solution makes an assay on this rabbit impossible. This rabbit proved to be consistently unsatisfactory and was discarded for purposes of assay.

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[*] A more frequent end to the usefulness of a rabbit is that its insensitive ear is eaten off by its fellows. The rabbits are thus best kept separate. Ears in which the great and posterior auricular nerves have been cut, but Arnold's nerve left intact, become sensitive after several months; the animals are then unsuitable for purposes of assay.

Results. We have now carried out more than a hundred assays of rabbit's kidneys for their renin content. Factors that influence the renin content of the rabbit's kidney and the significance of such variations will not be considered here. In this paper we are merely concerned with the results as far as they illustrate the accuracy and usefulness of the method of assay which we have described.

We therefore give in the accompanying table the results obtained by the two methods of extraction, and subsequent biological assay of the extracts, in a series of normal rabbits and in a series of rabbits which were interfered with (pathological rabbits). The renin content is expressed in units per g. kidney, the unit being that defined on p. 223.

The amount of renin in rabbit's kidney as assayed by the methods described in the text.

Description of rabbits.	Renin in units per g. kidney.	
	Alcohol method.	Globulin method.
Normal	3·94	5·0
	3·4	3·25
	2·7	2·8
	2·25	2·5
	2·15	1·85
	1·9	1·8
	1·66	2·5
	1·64	2·4
Pathological	0·54	0·92
	0·35	1·0
	0·15	0·30
	0·07	0·28

Comparing the results obtained by the two methods of extraction in individual kidneys, we find that the values obtained by the total globulin method are higher in 9 and lower in 3 instances than the results given by the alcohol method. In the normal group where the renin content is relatively high, the globulin result is never more than 50% above or 15% below the alcohol value. In the pathological group, where the renin content is low, the difference is greater, and in the last instance the globulin value is 4 times that obtained by the alcohol method.

In considering these results, two points should be borne in mind. In the first place the comparison of unknown and standard solutions on the rabbit's blood pressure involves a possible error of at least 25% in the assay of each extract. Some lack of uniformity would thus be anticipated. In the second place alcohol has already been shown to inactive renin, and higher values would thus be expected from the globulin than from the alcohol method of extraction. The results suggest that this action of alcohol introduces a greater source of error when the renin content is low than when it is high.

Nevertheless, when a variation in one direction of the renin content is shown by one of the methods of extraction, it is usual for the other to show a variation in the same direction. Thus, in the table the results are arranged in order of diminishing renin content as found by the alcohol method. With three exceptions, the results obtained by the globulin method fall in the same order. Thus it is probably safe to use one or other method of extraction in determining the influence of various factors on the renin content of the kidneys, provided this method is used throughout, and provided that the differences investigated are considerable. The alcohol method is the more convenient, but the total globulin method is probably the more accurate, particularly when the renin content is low.

Further improvement in the method should be possible as our knowledge of renin grows. But it is evident, from the few results we have given, that variations in renin content do occur, which are well within the present limits of estimation of the method, and it is possible that the study of such variations may assist in determining the role of renin in the organism.

SUMMARY.

1. A prolonged rise of blood pressure may be produced in the unanæsthetised rabbit by intravenous injection of extracts prepared from fresh kidney of several species, by methods described in the text. The active substance, " renin," is present in the cortex but not in the medulla of kidney. It has not been recovered from other tissues of the normal rabbit.

2. Renin is non-dialysable, and, in neutral solution, is destroyed at 60°C.. It is insoluble in alcohol, acetone, ether and chloroform. It is inactivated by alcohol and acetone at temperatures above 0°C. but is stable to alcohol at —10°C.. It is destroyed by strong acids and alkalies. It is precipitated by half saturation with ammonium sulphate and is distributed between the euglobulin and pseudoglobulin protein fractions, but chiefly in the latter. These facts seem to indicate that it is a protein.

3. The pressor action of renin is reduced or abolished by anæsthetics such as urethane, nembutal, and ether.

4. Renin may be assayed biologically. To act as a standard, a stable preparation of renin has been made by drying and powdering the residue

from rabbit's kidneys treated with alcohol. Solutions made from standard powder and from tissues to be assayed may be compared by using the blood pressure response of the unanæsthetised rabbit.

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FURTHER OBSERVATIONS ON FAMILIAL

PERIODIC PARALYSIS.

By E. N. ALLOTT and B. McARDLE.

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In a case of Dr. M. Walker's, one of us (E.N.A.) (16), found in 1935 a lowered serum potassium during attacks of familial periodic paralysis. In a subsequent publication (1) it was shown that the attacks could be relieved by the administration of potassium salts by mouth, and could be brought on by a variety of procedures, which had in common the effect of lowering the serum potassium. We have had the opportunity of confirming these findings in two new cases and have been able to extend the observations in certain directions.*

Case histories.

Case 1. The patient described and investigated in the previous paper. This patient has since died of pneumonia ; Dr. Walker has informed us that during his last illness he was nearly continuously in an attack of paralysis, but no chemical observations were made. Postmortem examination by Dr. P. D. Day showed to the naked eye nothing abnormal except the pneumonic changes. Histological examination has shown some so far unidentified droplets in the muscles : further histological studies are being made by Dr. Day.

Case 2. R.H., a painter, aged 28, was first admitted to Addenbrookes' Hospital under the care of Dr. Cole in November, 1936, and subsequently in February, 1937. One paternal uncle and one brother suffered from the complaint : his father, who died aged 63 had a few attacks : two brothers and two sisters are normal, and the patient has one normal son, aged 5. The patient had his first attack at the age of 13, when he became completely paralysed within 10 minutes, and remained paralysed for 15 hours. His

next attack was at the age of 15 and since then attacks have occurred once or twice a month, becoming more frequent till the age of 24 years, since when they have been somewhat less frequent. The day before an attack he gets a warning in the form of a pain in both arms. As in Case 1, the attacks usually occur at night. He wakes up at about 3 a.m. or 4 a.m. : the attack usually starts as a weakness of his quadriceps and spreads upwards and downwards from this point : the arms are usually affected late in the attack : the fingers first, then the shoulders, then the rest of the arm. The toes usually escape and he is able to move them throughout the attack. His abdominal and intercostal muscles, and the intrinsic muscles of the larynx, fauces and tongue are generally spared. The paralysis usually takes some hours before it reaches its maximum, and then passes off slowly. If he is able to get out of bed, he finds that he can often abort an attack by walking it off. There is no disturbance of sensation or loss of consciousness and no interference with sphincter control. He occasionally vomits during an attack. The most important predisposing factor seems to be emotion or excitement of any sort, though a heavy meal late at night is frequently followed by an attack. Between attacks he is found on physical examination to be a perfectly normal individual.

Case 3.   W.I., a labourer, aged 48, was admitted to the National Hospital, Queen Square, under the care of Dr. Symonds. His father, aged 84, suffers from the condition, as does one elder brother : his eldest brother is quite well : no other member of the family is known to suffer and the patient has one son, aged 9, who is quite normal. The attacks started at the age of 16 and until January, 1937, came on about once or twice a year, lasting about three days, and not inconveniencing him very much. As in both previous cases the attacks come on at night, and the patient awakes to find himself unable to change his position in bed. The legs seem to be affected on the whole a little earlier than the arms, and are usually later in recovering. Sometimes the paralysis also affects the trunk muscles, and there is difficulty in breathing, and the patient has not the strength to cough satisfactorily. Speech and swallowing are sometimes interfered with. Dr. M. J. McArdle has observed that muscles placed in extension manifest paralytic phenomena to a greater degree than those in relaxation, and further that placing a paralysed muscle in a relaxed position will sometimes permit some return of power. Since an attack in January, 1937, he has never made a complete recovery : he has never been really free from paralytic symptoms for more than two or three days and is generally in the stage of development or disappearance of an attack. Since this time he has been an invalid. This is the condition of the patient's brother, who has been in an institution for 8 years as he has never been free from attacks long enough to keep in employment. There are no very definite predisposing factors : exposure to wet or cold weather sometimes produces an attack : there is no regular warning, but sometimes he has unusual thirst the evening before an attack.

Action of insulin and glucose.

In the first case studied it was shown that attacks of paralysis, identical clinically with a natural attack, could be produced by the administration of glucose by mouth, by insulin or more consistently, by glucose and insulin together. Weakness developed when the serum potassium had fallen to about 12 mg. per 100 c.c. and a complete attack when the level had fallen to about 10 mg. per 100 c.c.. In both the new cases attacks could equally readily be produced with glucose + insulin, the changes in serum potassium being very similar to that previously observed, a typical experiment in each case being shown in Figs 1 and 2. In Case 2 recovery on the administration
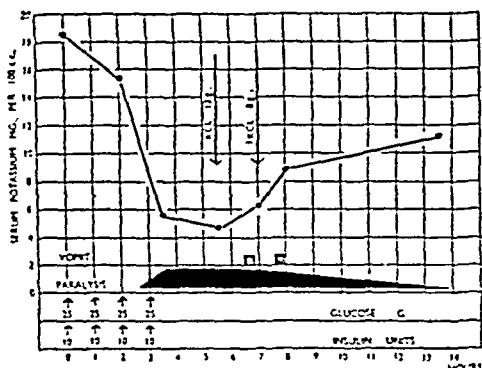

Fig. 1.
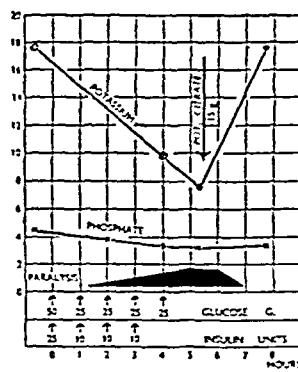
Fig. 2.

Fig. 1. A typical attack of paralysis produced in Case 2 by glucose and insulin.

Fig. 2. An attack produced by glucose and insulin in Case 3.

Top curve, serum potassium, mg. per 100 c.c.

Bottom curve, serum inorganic phosphate, mg. per 100 c.c.

of potassium chloride was prolonged, owing to most of the drug being vomited, but with the rise of serum potassium to 11·3 mg. per 100 c.c. power had nearly returned. In Case 3, the intermediate values did not enable one to give accurately the serum potassium level at which power had returned, but from other experiments it would appear to be much higher than that in Case 1.

Action of adrenaline.

It has been shown by D'Silva (4), (see also Schwarz (14)) that the administration of adrenaline to anæsthetised cats produces a very rapid rise in the serum potassium, followed by a rapid fall : the rise was as much as 12 mg. per 100 c.c. above the initial value, but within 4 mins the normal level was regained, and in 10 mins the serum potassium had fallen much below its original level. The effect of adrenaline has been investigated in all our three cases of familial paralysis and in three normal controls. In

each individual, the subcutaneous injection of 0·6 to 1 c.c. of 1 in 1,000 adrenaline has produced a fall in the serum potassium : on two occasions only* has there been any trace of the initial rise described by D'Silva, and in general there has been a fall from the beginning, even when blood was taken within 1 min. of injection. Keys (12) and Castleden (3) have made similar observations in normal subjects.

In a normal individual, the response to subcutaneous injection of 0·6 c.c. 1 in 1000 adrenaline has been a fall of some 3 to 4 mg. per 100 c.c. in serum potassium level, the effect being maximal in about half an hour, and the level then returning to normal in the course of the next one to two hours (Fig. 3). By repeating the adrenaline administration to a normal individual, 1 c.c. every 20 minutes until a total of 5 c.c. had been given,
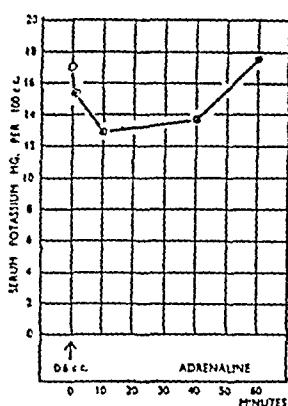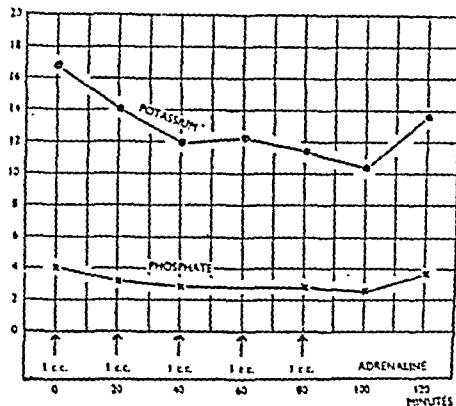

Fig. 3.   Fig. 4.

Fig. 3. Effect of a single dose of 0·6 c.c. adrenaline (1 in 1000) on the serum potassium of a normal individual.

Fig. 4. Effect of repeated injections of adrenaline (1 in 1000) on the serum potassium and phosphorus of a normal individual.

Top curve, serum potassium, mg. per 100 c.c.

Bottom curve, serum inorganic phosphate, mg. per 100 c.c.

it was possible to reduce the serum potassium to 10·5 mg. per 100 c.c. but no paralytic manifestations of any kind were produced (Fig. 4).

Two of the patients (Cases 1 and 3) responded normally to adrenaline and the serum potassium was not reduced to an extent sufficient to produce a definite attack of paralysis, although in Case 3 where symptoms came on usually at a higher potassium level than in the other two cases, there were mild symptoms. Potassium excretion in the urine during the adrenaline experiment on Case 3 is also shown in Fig. 5 : although the fall in serum

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* Both occurred in Case 1. In one instance the adrenaline (1 c.c.) was given when the patient was in an attack produced by insulin + glucose, and produced a momentary rise from 10·3 mg. per 100 c.c. to 22·0 mg.. In the other instance, the adrenaline (1 c.c.) was given while the patient was in his normal state, and produced a rise from 22·3 mg. per 100 c.c. to 25·3 mg. in 1½ min.. There seems no reason to doubt the accuracy of these analytical figures, but they stand out as exceptions to a much larger number of observations where there was a fall from the beginning.

potassium was not great, there was a very great diminution in potassium excretion during the period when the adrenaline was acting. In the remaining patient, (Case 2), however, the effect was very different. As in the observations with glucose + insulin, the initial fall was much greater than normal, and the serum potassium subsequently showed no tendency to return to normal but, on the contrary, continued to fall until the threshold necessary to provoke an attack was passed (Fig. 6). This mechanism could be consistently put into operation by the subcutaneous injection of some 0·6 c.c. of adrenaline (1 in 1000).

It is of interest that the patient in whom attacks were consistently brought on by adrenaline was one in whom the emotional factor played a
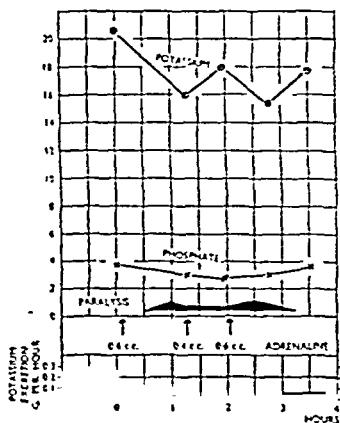

Fig. 5.   Fig. 6.
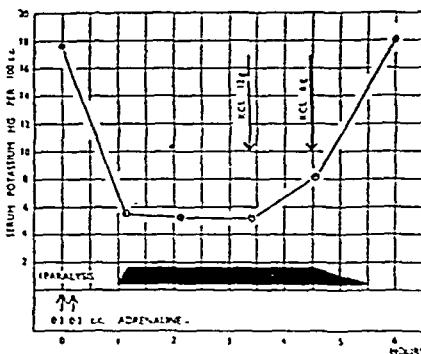
Fig. 5. The effect of repeated doses of adrenaline (1 in 1000) on the serum potassium and phosphorus in Case 3. The potassium excretion in the urine is also shown.

Top curve, serum potassium, mg. per 100 c.c.

Second curve, serum inorganic phosphate, mg. per 100 c.c.

Fig. 6. The effect of subcutaneous injection of adrenaline (1 in 1000) on serum potassium in Case 2.

large part in bringing on the attacks, whereas emotion was not so definitely associated with the onset of attacks in the other two cases.

Guttmann (8) has described a case of familial periodic paralysis in which also adrenaline regularly produced attacks, and in which the attacks, whether spontaneous or induced, could be terminated by the injection of pilocarpine. Pilocarpine in large doses (1/6 grain) was ineffective in terminating a spontaneous attack in Case 3, although producing much sweating and nausea. Guttman stated that in his case there was a very dry skin during attacks, and profuse perspiration at the end. In our cases, the skin was not noticeably dry during attacks, and there was not any obvious sweating at the end. Shinosaki (15) was able to produce attacks by means of adrenaline in only

3 out of 12 cases in which the drug was tried ; in no case was he able to produce an attack by pilocarpine : pilocarpine was not tried with a view to terminating an attack.

Adrenaline thus produces an effect on potassium metabolism similar to that observed in a spontaneous attack, namely, retention of potassium in the body, with fall of serum potassium (*see* Fig. 7). We therefore tried the effect of two other hormones (parathormone and eucortone) whose action is to accelerate the excretion of potassium, and which might be expected to lower serum potassium : in neither case was an attack of paralysis obtained, but the fall in serum potassium was small with eucortone, and absent with parathormone.

Parathormone.

Eighty units of parathormone were injected intravenously in Case 3. Apart from malaise some 3 hours after the injection, no symptoms were observed. As Table I shows, there was no significant change in the serum potassium.

TABLE I.

Excretion of potassium and phosphorus after parathormone injection (80 units intravenously at 2.35 p.m.).

Period.	URINE.			SERUM.	
				Potassium.	
	Volume c.c.	Potassium g.	Inorganic phosphate g.	mg. per 100 c.c.	Time.
11.0—12.0	38	0·10	0·027	—	—
12.0—1.0	18	0·07	0·025	—	—
1.0—2.30	150	0·09	0·054	18·4	2.35 p.m.
2.30—3.30	362	0·47	0·051	19·0	3.40 ,,
3.30—4.30	83	0·20	0·047	17·5	4.45 ,,
4.30—5.30	32	0·10	0·037	—	—
5.30—10.0	100	0·19	0·103	—	—

During the two hours following the injection, approximately 0·5 g. extra potassium was excreted : the extra water was approximately 350 c.c. which, if it is all extracellular water, should contain only some 0·07 g. potassium. The additional potassium must have come from some part of the tissues. The extra potassium was approximately the same in quantity as that found by Ellsworth and Nicholson (6) in normal subjects after the administration of 4 c.c. (80 units) of parathyroid extract. We have observed no difference in serum potassium level before and after operation in one case of parathyroid tumour with osteitis fibrosa.

Eucortone.

Little work has been done on the effect of suprarenal cortical extract on normal animals. Harrop and Thorn (9) have shown that cortical extract acts on normal dogs by slowing the excretion of sodium and chlorine and hastening that of potassium : no figures are given for its effect on serum potassium.

The intravenous injection of eucortone (concentrated preparation, 1 c.c. = 75 g. suprarenal cortex) in doses of 10 c.c. at 3 p.m. and 6 c.c. at 9 p.m. produced no significant change in potassium excretion during the ensuing 12 hours, and no symptoms of any kind. The changes in the serum are shown in Table II.

TABLE II.

	Serum potassium.	Serum inorg. phosphate.
3 p.m. Eucortone 10 c.c. i.v.	20·8	3·09
9 p.m. „ 6 c.c. i.v.	18·9	3·52
Midnight	16·4	4·06

(The values are given in mg. per 100 c.c.).

Potassium and phosphorus excretion in relation to attacks.

For a period of 5 weeks Case 3 was maintained on a diet of known potassium and phosphorus content. For the first fortnight the potassium intake was approximately 2·6 g. per day, for the remaining period, approximately 1·4 g.. The phosphorus intakes during the two periods were 1·09 g. and 0·71 g., respectively, expressed as phosphorus. Urine was collected in 12 hourly periods from 10 a.m. to 10 p.m. and 10 p.m. to 10 a.m.. Potassium, inorganic phosphate and creatinine were determined in these specimens. No attempt was made to allow for the amount of potassium excreted in the perspiration, which is about 0·15 g. per day according to Freyberg and Grant (7), or in the fæces.

The creatinine was determined as a check on the accuracy of collection. Creatinine output was sensibly constant, with a few irregularities not related to attacks, but was slightly higher during the period of low potassium intake (mean value 1·04 g. per day as against 0·92 g.). Normal creatinine excretion in familial periodic paralysis had previously been observed by Edsall and Means (5) and Kirk and Møller (13). The total excretion of creatinine was not appreciably different by night and by day, but that of potassium and phosphorus during the night period 10 p.m. to 10 a.m. was always much less, and usually about half that of the day output.

The excretion of potassium and phosphorus during 24 hourly periods is shown in Fig. 7. It can be clearly seen that the potassium excretion is

THE EFFECT OF ADRENALIN ON THE SERUM POTASSIUM LEVEL IN MAN.*

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DURING the study of a case of Familial Periodic Paralysis (1) it was shown that the attacks of paralysis were coincident with a fall of serum potassium level to about half the normal value, and that raising this to normal level by the oral administration of potassium chloride restored the power to the paralysed muscles. Attacks could be induced at will by any of three methods, namely, by giving sugar (by mouth) in large amounts, by injecting insulin, or by giving both sugar and insulin. Each of these procedures brought about a large fall of serum potassium level in the patient and a similar, though lesser, fall in control subjects ; this fall took place whether hypoglycæmia occurred or not. A fall in serum potassium following insulin injections given therapeutically to diabetics has been reported by Harrop and Benedict (5) and a fall of potassium and inorganic phosphorus in experimental animals after insulin injection by Briggs, Koechig, Doisy and Weber (2) and by Kerr (7).

It was suggested that the fall in serum potassium might be associated with the passage of sugar out of the blood into the muscles because that seemed the most obvious common feature in the actions of ingested sugar and injected insulin.

Schwarz (8) and D'Silva (4) described a rise in the serum potassium of rabbits and cats following intravenous doses of adrenalin. 1 mg. of adrenalin given intramuscularly to the patient during an attack of paralysis however, did not raise the serum potassium or alleviate the symptoms. The effect of a similar dose was therefore tried between attacks and was found to cause a fall of 8 mg. in serum potassium (Fig. 1). An attack of paralysis did not occur, presumably because the potassium had not fallen to the level usually associated with an attack in this patient. In other cases of periodic paralysis, however, the production of attacks by adrenalin has been reported by Shinosaki (9).
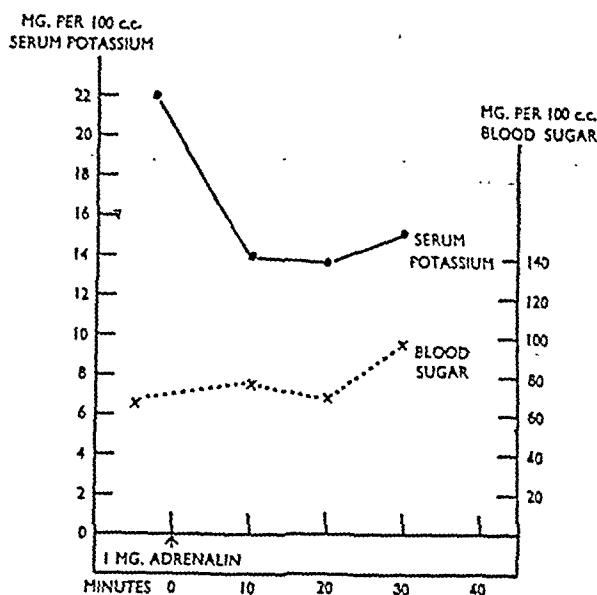
Fig. 1. Serum potassium and blood sugar in mg. per 100 c.c. of serum and blood respectively, estimated on venous blood before and at ten-minute intervals after giving an intramuscular injection of 1 mg. of adrenalin to the patient with Familial Periodic Paralysis, between attacks.
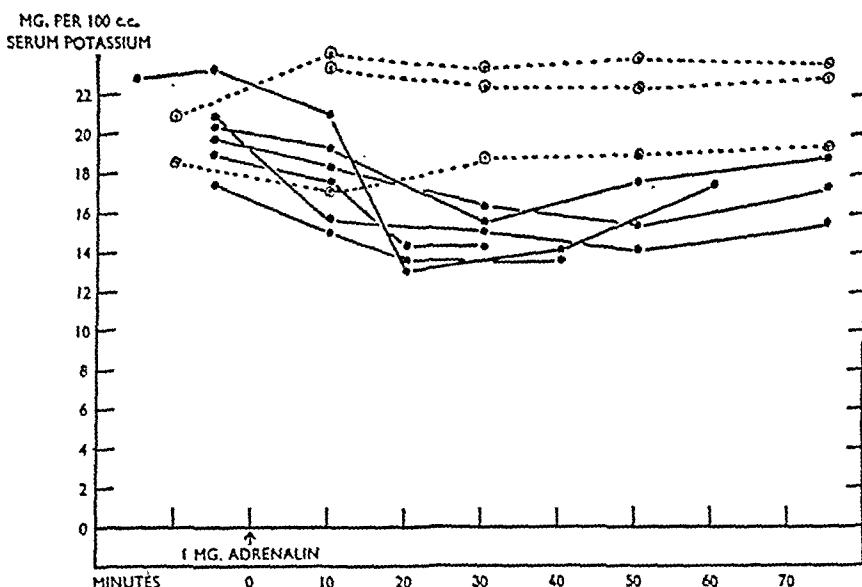
Fig. 2. The continuous lines show the effect of an intramuscular injection of 1 mg. of adrenalin on the serum potassium level in six subjects ten minutes and later after the injection. The dotted lines are control observations on three of the same subjects after an intramuscular injection of normal saline.

The response to adrenalin was therefore investigated in normal subjects. The first series of observations was made on four healthy men, aged 24, 28, 36 and 37, a man of 54 undergoing medical treatment for peptic ulcer

and a woman of 38 admitted to hospital for the investigation of functional dyspepsia. Serum potassium was estimated by the method of Jacobs and Hoffman (6). The serum potassium values at ten-minute intervals after an intramuscular injection of adrenalin are shown in Fig. 2. In all cases a fall occurred.

In his experiments on animals D'Silva (4) recorded a rapid rise of serum potassium to a maximum in about four minutes followed by an equally rapid fall to values a little below normal, after which a slow return to the resting level occurred. Although it might be expected that the intramuscular route, imperative in giving large doses of adrenalin to the human subject, would have delayed the time of response beyond that obtained by D'Silva who gave his doses intravenously, it was thought that by taking blood for serum potassium estimation at ten minutes after the injection a rise might have been missed between the injection and the first sample of blood. A
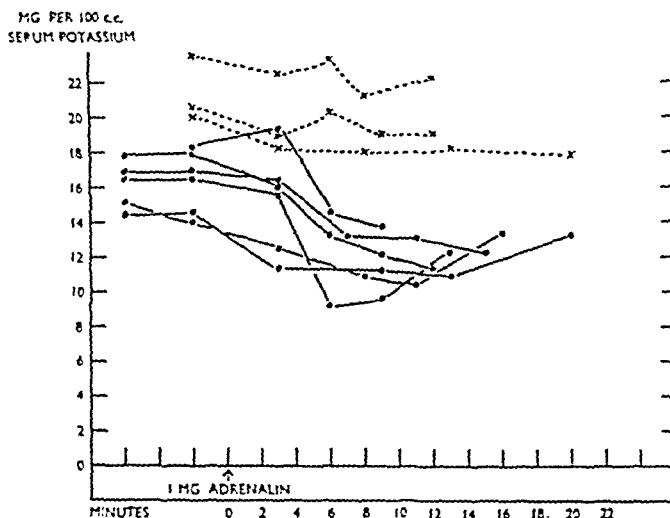

Fig. 3. The continuous lines show the effect of an intramuscular injection of 1 mg. of adrenalin on the serum potassium levels in six subjects at three minutes and later after the injection. The dotted lines are control observations after an intramuscular injection of normal saline.

second series of experiments was therefore performed on three men, aged 27, 41 and 54 and undergoing medical treatment for peptic ulcer, two men, aged 25 and 71, who were in hospital for the investigation of headaches, and a mentally deficient woman aged 28. The serum potassium values at three minute intervals after an intramuscular injection of adrenalin are shown in Fig. 3. With one exception, in which the blood obtained at three minutes after injection was haemolysed and therefore unreliable, all the curves show a fall, beginning at three minutes and afterwards continued.

During all these observations the blood pressure and the blood sugar were also recorded at frequent intervals ; but no constant correlation was

found between changes in either of these and the fall in serum potassium. In some cases the sugar curves were almost the mirror images of those for potassium, but in others the correspondence was not very close.

The average fall in serum potassium in the twelve observations was 5·3 mg. from an average resting level of 18·2 mg., and this occurred 10 to 20 minutes after the injection of adrenalin. This means that about 130 mg. of potassium was removed from the circulation during this period. If it were excreted by the kidneys, as seems possible, the rise in the potassium content of the urine should be measurable. To test this three excretion experiments were carried out on three normal men, aged 28, 36 and 37, in which both the serum potassium levels and urinary excretion of potassium were followed after an intramuscular injection of adrenalin and after a control intramuscular injection of normal saline. Table I shows that the injection of 1 mg. of adrenalin was followed by a fall in the excretion of potassium as well as by a fall in the serum potassium level. Renal excretion could therefore be excluded as a cause of the fall in serum potassium level. Harrop and Benedict (5) observed a similar fall in urinary potassium after therapeutic insulin injection.

TABLE I.

TIME.	EXPERIMENT 1.		EXPERIMENT 2.		EXPERIMENT 3.	
	Serum potassium, mg. per 100 c.c.	Urinary potassium, mg. per hour.	Serum potassium, mg. per 100 c.c.	Urinary potassium, mg. per hour.	Serum potassium, mg. per 100 c.c.	Urinary potassium, mg. per hour.
9.30	—	Bladder emptied	—	Bladder emptied	—	Bladder emptied
10.0	—	210(220)	—	72(221)	—	210(145)
10.30	—	198(264)	—	267(213)	—	231(302)
10.45	20·3	—	20·7(20·6)	—	20·1(18·5)	—
11.0	—	294(256)	—	314(290)	—	276(308)
Injection given immediately bladder was emptied at 11.0 a.m.						
11.10	19·5(23·3)	—	15·9(24·1)	—	18·4(16·8)	—
11.20	—	288(264)	—	241(261)	—	265(276)
11.30	15·6(22·1)	—	15·5(23·2)	—	16·4(18·7)	—
11·40	—	132(252)	—	61(256)	—	114(322)
11.50	17·6(22·1)	—	14·1(23·9)	—	15·8(18·8)	—
12.0	—	93(297)	—	54(198)	—	118(313)
12.15	18·5(22·3)	—	15·6(22·5)	—	17·4(18·6)	—
12.30	—	108(260)	—	58(291)	—	75(256)

The results of three excretion experiments on normal subjects who came up to the laboratory fasting and were kept semi-recumbent during the test. At 9.30 a.m. the bladder was emptied and 100 c.c. water drunk. This was repeated at half hour intervals up to 11.0, when 1 mg. of adrenalin was injected intramuscularly. 100 c.c. water continued to be taken at half hour intervals, but the bladder was emptied at 20, 40, 60, and 90 min., and samples of blood taken at 10, 30, 50, and 75 min. after the injection. Values in brackets were obtained on the same individuals after a control injection of normal saline under similar conditions.

The main conclusion from these experiments is that a fall of serum potassium level after adrenalin seems to be a regular phenomenon in the human subject.

Thus, there are three methods of lowering the serum potassium level, namely, injection of insulin, ingestion of sugar, and injection of adrenalin ; the first two cause an accelerated passage of sugar from the blood into the tissues but adrenalin does not, for it has been shown not to increase the arteriovenous sugar difference in man (3) and (10). It is therefore impossible to regard the fall in potassium level as related in all three cases to the passage of sugar from blood into tissues.

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UNILATERAL LOSS OF A BLOOD PRESSURE RAISING, PULSE ACCELERATING, REFLEX FROM VOLUNTARY MUSCLE DUE TO A LESION OF THE SPINAL CORD.

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It was demonstrated in an earlier paper (1) that the retention within voluntary muscles of the metabolites of muscular exercise causes an increase in the general blood pressure. This increase was attributed to a blood pressure raising reflex arising as a result of the chemical stimulation of nerve endings situated in the voluntary muscles, by substances which are formed or liberated in the muscles during exercise. The rise of blood pressure is elicited conveniently by arresting the circulation through an exercising limb or part of a limb by a sphygmomanometer cuff, thus preventing escape of the metabolites which are liberated during the exercise. During the actual performance of the exercise with ischæmic muscles the general blood pressure rises progressively. When this local muscular exercise ceases the blood pressure falls somewhat but nevertheless remains elevated well above the resting level for as long as the metabolites formed during the exercise are retained within the exercised limb. On removing the obstruction to the flow of blood through the limb, thus allowing the removal of accumulated metabolites, the general blood pressure falls rapidly to the normal level. To test further our conclusion (1) that this blood pressure increase is of reflex origin we decided to study the effects upon the blood pressure of local exercise with arrested circulation in a patient with complete loss of sensation below the knee in one of the legs. The observations made on this patient enabled us also to obtain further evidence that accumulation of the metabolites of muscular exercise in the voluntary muscle of the legs causes a reflex acceleration of the pulse (2).

The subject of our experiments is a stolid Egyptian subject aged 35 years.

Case history.

The patient complained of a deep painless ulcer, present for 15 years on the under surface of the heel of the right foot, and of loss of sensation in the right lower limb of about the same duration. The ulcer was roughly circular, about 1¼ inches in diameter and had never healed completely during the 15 years. It was shortly after the appearance of the ulcer that the patient first noticed that the right lower limb was becoming insensitive to heat, cold and pain.

Physical examination of the nervous system. Skin sensation was tested by the application of cotton wool, deep pricks with a sterile needle, ice and test tubes containing water at about 50°C.. Sensation from deeper structures was tested by movement of joints, heavy squeezing of muscles, a large vibrating tuning fork and by attempts to induce pain of the intermittent claudication type in muscles (3).

The feature of the case which concerns this investigation is total loss of all types of sensation in the right lower limb up to 4 inches above the knee with no sensory defect of the corresponding parts of the left lower limb. Complete anæsthesia of the skin extended also up the sides and back of the thigh to the gluteal region but strong cutaneous stimuli were felt on the anterior surface of the thigh to 4 inches above the patella. The cutaneous sensibility to heat and cold and pin pricks was diminished round the anus and in the lower part of the abdomen and in the upper part of the left thigh. Joint sense and vibration sense and appreciation of passive movements appeared to be entirely lacking in the right lower limb and normal in the left lower limb. All sensations above the umbilicus were appreciated in the normal way.

The knee jerk was absent on the right side and present on the left side. The ankle jerk was absent on both sides. The plantar response was absent on the right side and flexor on the left side. Muscle power was only slightly diminished on the right side and there was slight wasting of the muscles of the right lower limb. The patient stated, and it was evidently true, that he had no difficulty in walking and there was only a slight limp. Romberg's sign was negative. Physical examination of the nervous system revealed no other abnormalities. The Wassermann reaction was normal, the cerebro-spinal fluid was normal and a radiogram revealed no defect in the vertebral column or the sacral region. The case is unusual in that complete organic loss of sensation in one lower limb is associated with almost normal muscle power in both of the lower limbs. The principal lesion appears to be in the lumbo-sacral region of the spinal cord but apparently the damage extends up to about the 10th dorsal root. The lesion apparently developed in the course of two or three years and then remained stationary for 12 or 13 years.

Method.

The investigations were made with the patient at rest in the sitting posture. A large sphygmomanometer cuff was placed round the thigh of the left (normal) lower limb. The general blood pressure was measured at frequent intervals in an arm using a mercury manometer. Measurements were continued until the blood pressure had fallen to a steady level. The sphygmomanometer cuff round the left thigh was now inflated rapidly to well above the patient's systolic blood pressure in order to arrest the circulation through the left leg. A weight of about 12 Kg. was now rested upon the left knee and the patient was instructed to exercise the muscles of the leg below the knee by raising and lowering the heels while keeping the toes resting on the floor. The number of muscle efforts were counted and the exercise was continued until a moderate degree of pain of the intermittent claudication type was experienced in the muscles of the exercising limb (3). The exercise was then terminated but arrest of the circulation of blood through the limb was maintained by the sphygmomanometer cuff. Blood pressure measurements were made at frequent intervals throughout the period of exercise and for several minutes after the cessation of exercise. Finally the sphygmomanometer cuff was deflated and general blood pressure measurements were made as the blood flow returned to the leg and were

continued until removal of accumulated metabolites by the blood stream had restored the general blood pressure to normal. The experiment was then repeated on the right leg. The amount of exercise performed by the insensitive right leg in most experiments was the same as that performed in the control experiments by the left leg. In some experiments, however, the patient was induced to perform more exercise with the right leg than he had performed previously with the left leg.

In some experiments pulse rates were counted during the period of rest before exercise started, during the period of continued arrest of the leg circulation after exercise had ceased and after restoring the circulation through the leg.

Results and discussion.

Our experiments were designed to compare the effect upon the general blood pressure and pulse rate of exercise of a normal leg with that of a leg without sensation. In all experiments the local muscular exercise of the legs was performed during arrest of the blood circulation to the limb and circulatory arrest was maintained for some minutes after the cessation of muscular exercise. The results of typical experiments are shown in Fig. 1. Two main facts emerged.

(1). During the actual performance of exercise by the sensationless leg the general blood pressure and the pulse rate increased to about the same extent as they increased during the performance of the same exercise by the normal leg. It is well known that at the onset of general muscular exercise there is a rapid rise of the blood pressure and pulse rate which takes place at once when the exercise starts. It is usually thought that such sudden circulatory changes cannot be explained by the liberation of metabolites during the exercise. Two other explanations have been suggested. (*a*) That these sudden circulatory changes are reflex in origin and are caused by afferent nerve impulses discharged from voluntary muscle whenever it undergoes contraction. (*b*) That the sudden circulatory changes are the result of the mental processes which initiate exercise. It has been suggested, for example, that the discharge of nerve impulses from the motor cortex may lead to activation not only of the motor neurones but also of the medullary and other vital centres.

In a previous communication (1) we referred to the fact that not only *general* but also *local* muscular exercise involving but slight muscular exertion was accompanied by prompt increase of blood pressure and pulse rate. In our present experiments with local muscular exercise the start of exercise was marked by a prompt rise of the blood pressure (Fig. 1, Protocol 1) and pulse rate (Protocol 1) not only in the experiments where exercise was performed by the normal leg but also when muscular exercise was confined to the insensitive parts of the right leg. Since the nerve pathway from the exercised parts of the right leg is interrupted completely, it follows that this

prompt increase in blood pressure and pulse rate at the onset of exercise cannot be attributed to nerve impulses leaving the exercising muscles. Furthermore, the increases of blood pressure and pulse rate are not caused by exercise metabolites leaving the exercised limb because the arrest of the
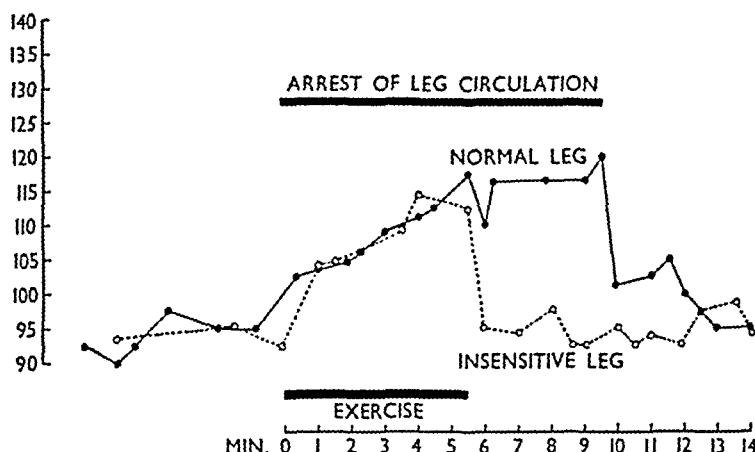

Fig. 1. Experiment showing loss of the blood pressure raising reflex from muscle in the insensitive leg and its presence in the normal leg of a subject with a spinal cord lesion.

blood circulation through the limb, both during and after the exercise, prevents such metabolites from entering the general circulation. Therefore the increase in blood pressure and pulse rate must be due to causes which originate outside the limb concerned. While lacking direct evidence we think the explanation most consistent with our results is that during the performance of local muscular exercise the associated cerebral events which lead to the discharge of nerve impulses down the motor nerves also cause nervous stimulation of the cell groups which control the level of the blood pressure and the rate of the pulse. The rapid fall of the blood pressure and pulse rate to their resting values, at the end of an exercise performed *by the insensitive* leg, also suggests a central nervous rather than a humoral cause for the blood pressure and pulse rate increases.

(2). When exercise of the normal leg or of either forearm was performed during ischæmia the general blood pressure and the pulse rate were raised above the normal level and on cessation of the exercise they remained elevated for as long as the circulation of blood through the exercised limb remained arrested (Fig. 1, Protocol 1). When, however, exercise was performed by the leg which was without sensation the blood pressure and pulse rate were raised during the actual performance of exercise but fell immediately to normal on cessation of the muscular exercise and remained at the normal level even though the circulation through the exercised muscles remained arrested. (Fig. 1, Protocol 1). Arrest of the blood circulation during and following the performance of local muscular exercise had led, in all our previous experiments on over 100 normal subjects, to a sustained rise of the blood pressure (1) and usually in the case of the legs of pulse rate (2).

PROTOCOL I.

In the following experiments the effect of exercise during ischæmia was compared in the two legs under conditions which were made as nearly as possible identical. The exercise which was performed separately by each of the legs consisted in raising a 12 Kg. weight up and down through a distance of about 2 inches 147 times. Separate experiments were made one on the healthy leg and one on the leg which was lacking in sensation. The results of the two experiments are set out side by side.

Time in min.	Experiment 1. Healthy leg.		Experiment 2. Insensitive leg.	
	B. P.	Pulse rate.	B. P.	Pulse rate.
	87/68		88/70	
	87/70		91/72	88
	87/67	86	89/69	88
	Circulation in leg arrested		Circulation in leg arrested	
	87/71		89/69	
0	Exercise starts		Exercise starts	
			105/78	98
1.30	96/73	90		
2.0	98/78			
2.30			108/80	98
3.0	99/81	92		
3.30	103/82	96	110/84	
4.0	108/84			
4.30	109/86	96	110/84	98
4.30	Exercise ends		Exercise ends	
5.30		98		88
6.0			89/73	88
6.30	108/84	101		
7.30	108/84	102	89/72	89
8.30	110/93	102	88/67	88
9.30	112/95		89/70	
9.35	Circulation restored		Circulation restored	
10.0	93/73		89/69	
10.30		96		90
11.0	94/72		91/70	
11.30	90/70	92		88
12.0	90/71		89/67	
12.30		88		88
13.0		86		
14.0		86	89/70	

In previous papers it was shown that the rise of blood pressure and pulse acceleration which occur when exercise of a leg is performed during ischæmia are explicable as reflex effects resulting from the accumulation of

exercise metabolites in muscles. The present observations confirm this view and observations with the blood pressure raising reflex similar to those described in this paper have been made in eight other neurological cases.

CONCLUSIONS.

Observations on a patient with complete loss of sensation in the right leg below the knee confirm the reflex origin of the increases in blood pressure and pulse rate which occur when the metabolites of exercise accumulate in the muscles of a normally innervated leg.

The presence or absence of these reflexes affords an objective method of studying the sensory disorders of muscle.

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OBSERVATIONS IN MAN CONCERNING THE EFFECTS OF DIFFERENT TYPES OF SENSORY STIMULATION UPON THE BLOOD PRESSURE.

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EXERCISE of the forearm muscles performed during arrest of the circulation of blood to a forearm leads to a rise in the general blood pressure (1) as well as to production of pain in the exercised muscles (3). Both the rise in the blood pressure and the pain persist after cessation of the exercise for as long as the circulation to the previously exercised muscles remains arrested. The magnitude of the persisting blood pressure rise and the severity of the persisting pain depend upon the amount of exercise performed during the ischæmia (1). Alam and Smirk showed that the rise of blood pressure produced by exercise of ischæmic muscles is reflex in origin and that a definite rise in blood pressure is obtained in many instances before the appearance of pain or of discomfort in the exercised muscles.

We have been performing many experiments in which the blood pressure has been raised by this reflex from exercised muscles and also by the immersion of an arm in water at 4°C. (2) and as pain was present to varying degrees in most of our experiments we undertook the present investigation to study the relationship between pain of various types and blood pressure increase.

Method.

We have examined the changes in the blood pressure and the pain produced by strong faradic stimulation of one or more fingers and by exercising the forearm muscles during ischæmia. The exercise consisted in alternately squeezing and releasing the rubber bulb of a sphygmomanometer. The circulation to the exercising forearm was arrested by means of a sphygmomanometer cuff applied round the upper arm, the cuff being inflated to well above the systolic blood pressure of the subject. The severity of the pain produced in this way and the degree of increase in the general blood pressure were controlled by altering the number of contractions

TABLE I.

Experiments in which equal blood pressure increases are produced by different types of sensory stimulation.

	Type of stimulation*	Maximum rise in B.P. (mm. Hg). Syst.	Diast.	Comments of subjects on degree of pain present at time when B.P. was measured.
SUBJECT 1.				
	A	26	19	Worse than B or C pain; almost intolerable.
Expt. 1	B	26	15	Less than A and worse than C. bearable.
	C	28	23	Definitely less than A or B and easily tolerated.
	A	24	18	More than B or C.
Expt. 2	B	25	14	Less than A and more than C.
	C	27	17	Definitely less than A or B and easily tolerated.
SUBJECT 2.				
	A	13	15	Fairly severe.
Expt. 1	B	13		Worse than A.
	C	18	19	Much less than A or B.
	A	17	14	Nearly intolerable.
Expt. 2	B	13		Less than A.
	C	13	6	Moderate pain. Less than A and B.
SUBJECT 3.				
	A	24	15	Greater than B.
Expt. 1	B	24	7	Greater than C.
	C	24	18	Definitely less than A or B.
Expt. 2	A	22	18	
	C	22	16	Less than A.
Expt. 3	A	37	14	Great pain.
	C	39	16	Worse than A.
SUBJECT 4.				
Expt. 1	A	10	—	Very severe.
	C	20	10	Slightly less than A.
Expt. 2	A	15	21	Very severe.
	C	16	20	Much less than A.
Expt. 3	A	33	29	Almost intolerable.
	C	35	30	Much less than A.

TABLE I.—*continued.*

	Type of stimulation.	Maximum rise in B.P. (mm. Hg). Syst.	Diast.	Comments of subjects on degree of pain present at time when B.P. was measured.
SUBJECT 5.				
Expt. 1	A	13	16	Moderately severe.
	B	16	22	Less than A.
	C	16	17	Definitely less than A or B.
SUBJECT 6.				
Expt. 1	A	13	16	Fairly severe.
	B	13	10	About equals A.
	C	14	8.	Less than A or B. Only slightly painful.
Expt. 2	B	19	22	Very severe.
	C	22	24	Much less pain.
SUBJECT 7.				
	A	24	—	
	C	23	30	Much less.
SUBJECT 8.				
	A	16	13	
	C	16	15	Less than A.

*A = electrical stimulation of a phalanx by the point of a copper wire electrode.
 B = electrical stimulation of three fingers immersed in saline.
 C = exercise of an ischæmic forearm.

performed. The raised blood pressures were measured after cessation of the exercise but with the circulation through the previously exercised arm arrested. The general blood pressure was measured on the unexercised arm. Electrical stimulation of the terminal phalanx of a finger was produced by applying the tip of a copper wire electrode to the bulb of a finger. The subject's forearm rested on the second electrode, a pad of cotton wool moistened with saline and supported upon a piece of zinc sheet. These two electrodes were connected to the secondary winding of an induction coil, the primary of the coil being connected to a series of accumulators. The strength of the electrical stimulation was adjusted by altering the distance between the primary and secondary coils. When stimulation of the three fingers was desired the same device was used but the wire which had been applied directly to the finger tip was dipped instead into the beaker of saline in which the three fingers were immersed. The blood pressure was measured by a mercury manometer.

and 12 mm. Hg. in the diastolic pressure. There was no change in the blood pressure from the electrical stimulation.

Our experiments show clearly that the degree of increase in the blood pressure depends upon the nature of the peripheral stimulus and is not an expression of the pain to which the stimulus gives rise. The rise of blood pressure from exercise of one forearm with arrested circulation usually is distinctly greater than that produced by exercise of both legs with arrested circulation (1) even when the degree of pain in the legs is greater than the pain arising from the forearm. Hence blood pressure increases do not depend merely upon the number of nerve fibres stimulated.

SUMMARY.

1. Some sensory stimuli cause much pain but little rise of blood pressure, others cause less pain but much greater increases of blood pressure.

2. Reflex increase of the blood pressure may be set up by nerve impulses which arise from voluntary muscle but which do not cause pain or discomfort. Either the blood pressure raising reflex from muscle is carried by nerve fibres other than those which carry the sensation of pain, or alternatively pain fibres from muscle may be stimulated in such a way that they cause a reflex increase of blood pressure without causing pain or any other definite sensation.

3. The reflex rise in the blood pressure which occurs when exercise metabolites accumulate in the voluntary muscles is not due mainly to the pain produced.

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BLOOD PRESSURE RAISING REFLEXES IN HEALTH, ESSENTIAL HYPERTENSION, AND RENAL HYPERTENSION.

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THE observations here recorded are concerned with increases of the blood pressure produced reflexly in hospital patients and in normal subjects with the object of studying the nervous control of the blood pressure in health and in hypertension. The effects of two different blood pressure raising reflexes have been investigated. One of the two stimuli used by us is cold applied to the skin of a hand and forearm. The effects of immersing one hand in water at 4°C. upon the general systemic blood pressure were described by Hines and Brown (8, 9) and by Pickering and Kissin (11). Hines and Brown found that immersion caused larger increases of blood pressure in patients with essential hypertension than in healthy subjects, but in a few of the apparently healthy subjects the increases of blood pressure were unusually large. In their experience those normal subjects whose blood pressure increased excessively, usually had near relatives who suffered from high blood pressure. Hines and Brown concluded that subjects who were apparently normal but had highly reactive blood pressures should be regarded as potential cases of essential hypertension. The average increase of the blood pressure is said to be greater in old than in young subjects (11). On comparing the effects of cold on the blood pressure in essential hypertension cases and in normal subjects of the same age group Pickering and Kissin (11) found that the blood pressure increases were not abnormally high in their cases of essential hypertension.

Method.

Our observations on the reflex effect of cold were performed in much the same way as those of Hines and Brown and Pickering and Kissin but we obtained relatively large blood pressure increases probably because not only the hand but also the lower 2/3 of the forearm was immersed in the cold water, and the circulation to the limb arrested. Circulatory arrest during immersion causes the arm temperature to fall more rapidly than when

the circulation is free, and has the further advantage that the degree of cooling is independent of the rate of blood flow through the arm. Under such conditions the actual stimulus is of constant intensity in all experiments, but its effectiveness varies in different subjects according to their sensitivity to cold.

The other method which we have used to raise the blood pressure reflexly is exercise of an ischæmic limb as described recently by Alam and Smirk (1). This blood pressure raising reflex appears to be one of the natural defensive mechanisms of the body designed to increase the blood flow through fatigued muscles by raising the general blood pressure; it is caused by nerve impulses leaving the exercised limb as a result of the accumulation of exercise metabolites and persists after exercise ceases provided the circulation remains arrested. This paper is concerned with the rise of blood pressure which persists after exercise ceases.

The exercise consisted of repeatedly squeezing the bulb of a sphygmomanometer in the hand or lifting and lowering a weight during local arrest of the blood circulation by means of an inflated sphygmomanometer cuff placed round the upper arm.

We instructed our subjects to continue the exercise until pain (10) becomes almost intolerable. With intelligent and co-operative subjects, we find that the rise of blood pressure obtained in this way is a few mm. less than the maximum blood pressure elicited by continuing the exercise until pain is intolerable. Thus the experiments with exercise are designed to raise the blood pressure in every case to just below the highest level to which it can be raised by the use of the reflex. The blood pressures were measured after the cessation of exercise but while the circulation through the exercised limb remained arrested. Measurements of blood pressure were made at $\frac{1}{2}$ to 1 minute intervals and usually for 5 or more minutes after the cessation of exercise. The blood pressure recorded is the highest blood pressure measured not less than 40 seconds after the cessation of the exercise. The levels of the blood pressures at rest and the effects of the reflexes upon the blood pressure were measured with the subjects in the sitting posture.

The subjects selected for study were normal subjects and patients with essential or renal hypertension. The normal subjects were, with one exception, of Egyptian nationality and all the patients were Egyptians. Some of the normal subjects were from the labouring classes, others were medical students and doctors. The patients with essential hypertension were without any defect in the excretory functions of the kidney; most had no trace of albumin in the urine and the few who had a trace of albuminuria gave urea concentrations of not less than 2·5%. In a few of the cases of essential hypertension the level of the systolic blood pressure after rest and repeated measurements of the blood pressure fell to between 140 and 150 mm. of mercury. The resting blood pressures of normal Egyptians have been found to be 10-30 mm. lower than the average blood pressures which are recorded for Europeans and Americans resident in their

own countries. It seems justifiable therefore to regard these pressures as abnormally high for Egyptian nationals. The diagnosis of such cases as essential hypertension seems justified also because their systolic blood pressures only fell below 155 mm. on resting for half an hour in the sitting posture and after making repeated measurements of the blood pressure. The systolic blood pressures measured after a short period of rest were over 155 mm. and usually over 165 mm.. The patients with renal hypertension were all definite cases with marked impairment of the capacity to concentrate urea and with strong clinical evidence that the renal lesion was not secondary to the hypertension. None of the cases had congestive heart failure.
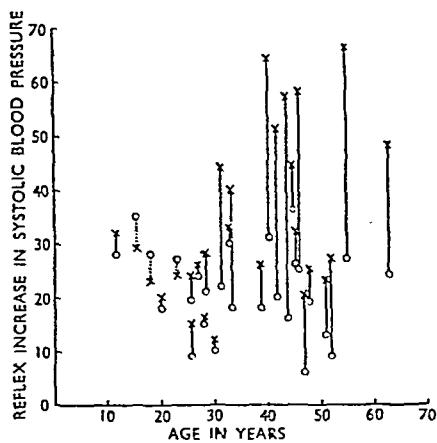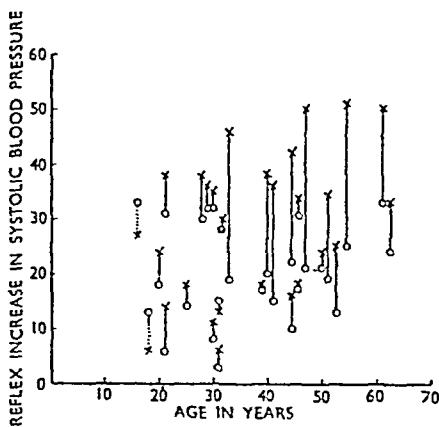

Fig. 1. Fig. 2.

Fig. 1. The effect of the reflex from muscle upon the blood pressure at different ages. Crosses are systolic pressure, circles are diastolic pressure increases.

Fig. 2. The effect of the local application of cold upon the blood pressure at different ages. Crosses are systolic pressure, circles are diastolic pressure increases.

Results.

1. The effect of age on the responses of normal subjects.

(a) *Experiments with the reflex from voluntary muscle.* The results obtained are set out in Fig. 1. The increases of systolic blood pressure range from 12 to 16 mm. of mercury and of diastolic pressure from 6 to 36 mm. of mercury. Large increases of systolic blood pressure in response to the reflex are much commoner in normal subjects over the age of 40 than in younger individuals. The increases of diastolic pressure however, are about the same in the old as in the young subjects. It follows that the reflex from voluntary muscle causes the greatest increases of the pulse pressure in the group of older subjects. Occasionally in the group of younger subjects diastolic pressure is raised more than the systolic. This observation gains interest if one recollects that the elasticity of the larger arteries decreases

with advancing age (5). This decrease with age in the elasticity of the arteries might in fact, explain our observation that blood pressure raising reflexes cause greater rises of the systolic blood pressure in old than in young subjects.

(*b*) *Experiments with the reflex from cooled skin.* The immersion of a hand and forearm in water at 4°C. led, in this series of subjects, to increases in the systolic blood pressure of 6-51 mm. of mercury and in the diastolic pressure of 3-33 mm. of mercury (Fig. 2). The age of the subjects had the same influence upon the reaction of the blood pressure to cold as it had on the reaction to the accumulation of exercise metabolites in muscle. The fact that larger increases in systolic blood pressure are encountered in the older subjects is in agreement with the observations of Pickering and Kissin (11).

As most patients with hypertension are over 40 years of age it follows that the reactions to blood pressure raising reflexes of such patients with essential hypertension should not be compared with the reactions of normal human subjects from a different age group.

2. *Responses to blood pressure raising reflexes in essential and in renal hypertension cases.*

The effects of the reflex from voluntary muscle upon the systolic blood pressures of normal controls, essential hypertension and renal hypertension cases are set out in Fig. 3. The effects of the immersion of a hand and the lower 2/3 of the forearm in water at 4°C. are to be seen in Fig. 4. The normal subjects used as controls were over the age of 40, i.e., of the same age group as the patients with essential hypertension; the patients with renal hypertension were also with one exception, above the age of 40.

It is evident that the increases of the blood pressure in response to both reflexes are less in the cases of hypertension due to chronic nephritis than in either the normal subjects or in patients with essential hypertension. The *average* rise in the blood pressure due to the reflexes is greater in the patients with essential hypertension than in the normal subjects of the same age group. Hines and Brown observed a greater difference than we did between the blood pressure increases in essential hypertension cases and normal subjects in response to the application of cold; Pickering and Kissin, however, found no difference between the blood pressure increases of normal and essential hypertension subjects. The difference between the results of Pickering and Kissin and of Hines and Brown may be explained partly by the fact that Pickering and Kissin had as controls subjects who were of the same age group as the hypertension cases, whereas Hines and Brown apparently regarded the age of the subjects as having little effect on the reactivity of the blood pressure to cold (9). Our results show that there is a very definite relationship between age and reactivity of the blood pressure and when the results of our experiments on hypertension cases are compared with the results we obtained on younger controls a difference of the same order as that observed by Hines and Brown is observed.

Our results were obtained under very similar conditions to those of Pickering and Kissin and we are unable to account satisfactorily for the difference between our results. We desire to emphasise, however, that care must be taken that the blood pressures of the essential hypertension cases
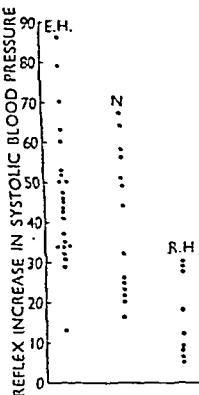

Fig. 3.
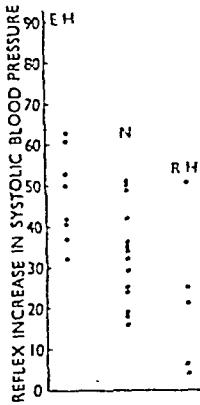
Fig. 4.

Fig. 3. The effect of the reflex from muscle upon the systolic blood pressure in essential hypertension, renal hypertension and in healthy subjects. In this and subsequent figures E.H. = essential hypertension, R.H. = renal hypertension, N. = normal subject.

Fig. 4. The effect of the local application of cold to a hand and forearm upon the systolic blood pressure in essential hypertension, renal hypertension and in healthy subjects.
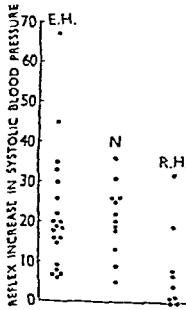

Fig. 5.
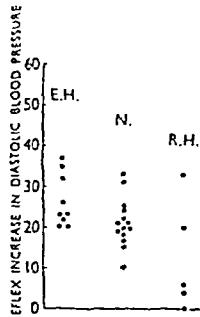
Fig. 6.

Fig. 5. The effect of the reflex from muscle upon the diastolic blood pressure in essential hypertension, renal hypertension and in health.

Fig. 6. The effect of the local application of cold to a hand and forearm on the diastolic blood pressure in essential hypertension, renal hypertension and in healthy subjects.

have had time to return to their true resting level before the blood pressure raising stimulus is applied, otherwise the full extent of the rise of blood pressure in response to a stimulus may not be realised. Rest and habituation

to the presence of a medical man and to the procedure of blood pressure measurement will lower the blood pressure both in normal and in essential hypertension subjects. The extent to which the blood pressure falls with mental and physical rest appears to be greater in the patients with essential hypertension than in healthy subjects.

In Figs. 5 and 6 the reflex increases in the diastolic pressures of normal subjects, essential hypertension and renal hypertension patients which were obtained in the above experiments are recorded. The increases are least in the cases of renal hypertension. Cold appears to raise the diastolic pressure to a greater degree in the patients with essential hypertension than in the normal subjects of the same age group.

Our observations are consistent with the view that the blood pressure is much more labile in essential than in renal hypertension.

Other examples of the lability of the blood pressure in essential hypertension are provided by the work of Barath (3) who observed that the rises of blood pressure resulting from emotional stimuli are greater in essential hypertension cases than in healthy subjects. Barath (3), Eppinger and Kisch (6) and Eppinger and Schwarz (7), Bauer and Neuburger (4), and Raab (12) found that the performance of controlled muscular work raises the blood pressure to a greater degree in essential hypertension than in health. Such observations indicate that the blood pressure raising actions of ordinary daily activities, mental, physical and emotional, have usually a much greater effect upon the blood pressures of patients with essential hypertension than upon the blood pressures of normal subjects.

Discussion.

It has been suggested by Raab (12) that the high blood pressure of essential hypertension is due mainly to a high degree of reactivity of the vasomotor system. Our observations indicate, however, that while a high reactivity of the blood pressure to blood pressure raising reflexes is more common in cases of essential hypertension than in normal subjects, yet in many cases of hypertension the blood pressure is no more reactive than it is in some normal subjects. This observation alone shows that high reactivity of the blood pressure to blood pressure raising stimuli cannot, by itself, explain a high level of the resting blood pressure. This conclusion is strengthened by a study in normal subjects of the relationship between the degree of reactivity of the blood pressure to blood pressure raising reflexes and the level of the resting blood pressure ; the resting blood pressure level is no higher in the more reactive than in the less reactive normal subjects. The average systolic blood pressure at rest of 12 normal subjects whose blood pressures were raised 40 to 60 mm. by the reflexes was 88·5, and 14 normal subjects whose blood pressures were raised by 0 to 20 mm. was 97·0. We conclude that a high degree of reactivity to the two blood pressure raising reflexes which we have studied is not associated with a higher level of the resting blood pressure in normal subjects. The average level of the resting

blood pressure in our more reactive patients with essential hypertension did not differ appreciably from the level of the resting blood pressure in the less reactive patients. The averages of the systolic blood pressures *at rest* in the patients with essential hypertension were 169 for those giving reflex increases of 40 mm. or more, and 171 for those giving reflex increases of less than 40 mm.

It is possible that a high degree of reactivity of the blood pressure to blood pressure reflexes may express itself in daily life by abnormally strong and frequent blood pressure variations and it is even possible that this may lead sometimes to the development of permanent hypertension. We desire, however, to stress the fact that the average of the blood pressures *at rest* of subjects with a high degree of reactivity to the blood pressure raising reflexes is no greater than the average blood pressure of subjects with a low degree of reactivity.

In so far as a high average level of the blood pressure is likely to increase the liability to congestive heart failure and to cerebral hæmorrhage one must regard the high reactivity of the blood pressure as of practical importance in essential hypertension since it appears to be responsible for exaggerating the rises of blood pressure which take place normally with emotional or physical activity and must be in part responsible for setting the daily as distinct from the resting level of the blood pressure at a high level.

It was claimed by Hines and Brown that apparently normal subjects who have unusually large increases in blood pressure in response to the application of cold to the skin should be regarded as potential cases of essential hypertension. The incidence of high reactivity in our normal subjects so far exceeds the incidence of high blood pressure in Egyptian hospital practice that we are confident that essential hypertension does not usually develop subsequently in healthy subjects with a high reflex reactivity of the blood pressure ; there remains for consideration the possibility that the incidence of essential hypertension may be higher than normal in these subjects.

It was noted in the course of our observations that in normal subjects the blood pressure increases elicited by the muscle reflex *from the arm* gave rise to either no appreciable change in the pulse rate or to a slight slowing of the pulse (2). In the patients with essential hypertension the pulse rate was usually increased when the blood pressure was raised reflexly in the same way. It has long been noted that the high blood pressure of essential hypertension is not associated with the abnormal slowness of the pulse which would be expected from Marey's law. Attempts to explain this absence of slowing of the pulse have been made by postulating adaptation or inactivity of the depressor mechanisms of the carotid sinus and aortic arch. Our observations show that in many cases of essential hypertension the usual relationship between pulse rate and blood pressure is reversed. This reversal would be of considerable interest if it could be shown to result from a change in the reactions of the depressor mechanisms.

As far as the writer is aware, these observations have not been repeated on man, although considerable interest has been shown in the effects of sympathectomy on angina pectoris. Those described in this paper were carried out on patients who were subjected to the operation of sympathectomy for some non-cardiac cause. The operations were performed (with one exception in which Mr. Philip Hawe was responsible) by Mr. J. Bagot Oldham, whose patience and willing co-operation was largely responsible for their success. The cases chosen included patients suffering from Raynaud's disease, excessive sweating and retinitis pigmentosa. In the cases of Raynaud's disease a cervico-dorsal ganglionectomy was performed, in which the inferior cervical ganglion was extirpated. In other cases the superior cervical ganglion only was resected. The interval between the operation on one side and that on the other varied from a week to a few months. Stimulation of the sympathetic ganglion was carried out either by inter-. mittent pressure with Spencer Wells forceps or by a weak faradic current. After stimulation, the sympathetic ganglion was extirpated.

Electrocardiographic records* were made before anæsthesia and during anæsthesia before operation, in order to exclude the effects on the electro-cardiogram of the anæsthetic. This proved to be a necessary precaution as not infrequently a definite change resulted from anæsthesia.

Further records were taken during stimulation, and 15-30 minutes after, and then at longer intervals of hours or days after the operation was complete. As the operations were conducted at different hospitals it was not possible to use the same electrocardiograph, though the majority of the records were taken with a Cambridge portable instrument. Eight cases in all were investigated, but two proved unsatisfactory for various reasons.

Results.

Stimulation of the right sympathetic. The most remarkable changes are to be observed after stimulation of the right sympathetic and the results are in many ways comparable with those of Rothberger and Winterberg working on dogs. The changes are well seen in the accompanying cardio-grams taken from the case which showed them in the highest degree. Other cases, however, showed similar changes in a lesser form. These changes consist of diminution in the amplitude of the R wave, and increase in the amplitude of the T wave. Sometimes both variations were found in the same record, sometimes only one. The changes lasted only for a few minutes after the stimulus ceased. The negativity of the T wave on stimulation of the right sympathetic, described by Otto, was not observed in the present series.

Stimulation of the left sympathetic. This produced no such variation in the form of the cardiogram; indeed the results were often negative.

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* I wish to thank Mr. E. Caldwell for his careful work in connection with the electro-cardiograms.

It was, however, not uncommon to find auricular extrasystoles, or evidence of shifting of the pacemaker to the junctional tissues ; but the results were inconstant and not infrequently similar effects were caused by anæsthesia alone.

Extirpation of the sympathetic on one or both sides caused no alteration in the form of the electrocardiogram, which resembled in all respects the preoperative record.

The alteration in the form of the electrocardiogram by sympathetic stimulation suggests that accelerator and vagus tone may be partly responsible for the form of the normal electrocardiogram, and variations in the preponderance of the two nerves may be responsible for some of the types of record seen in healthy persons.

CONCLUSION.

In man, resection of the cervical sympathetic ganglia has no effect on the form of the electrocardiogram. Stimulation of the right sympathetic may change the form conspicuously, decreasing R, or increasing T, or both.

Stimulation of the left sympathetic produces no constant changes.

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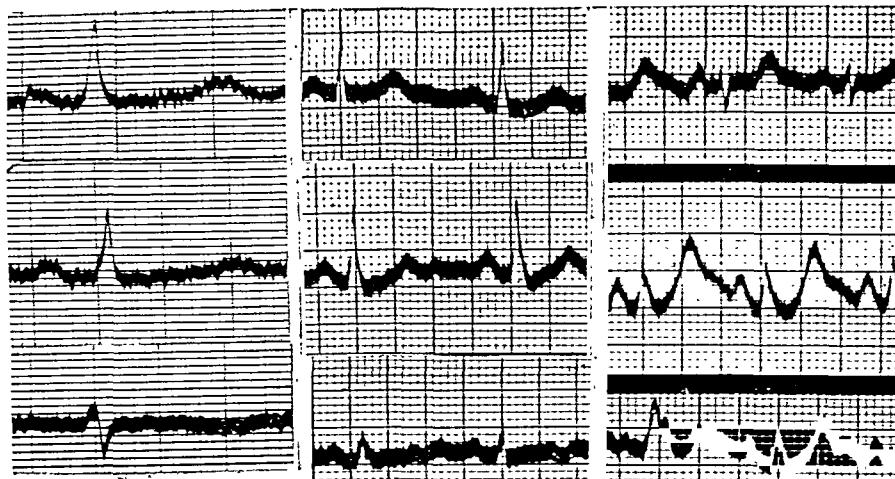
Fig. 2. Record taken at rest before operation 10/5/33. Shows left ventricular preponderance.

Fig. 3. Record taken during anæsthesia, before surgical interference 11/5/33. Slight change in *QRS* complex of lead III.

Fig. 4. Record taken during stimulation of right stellate ganglion 11/5/33. Note increased amplitude of *T* waves and diminished *R* waves. Also tendency to R.V.—and slight increase in heart rate.
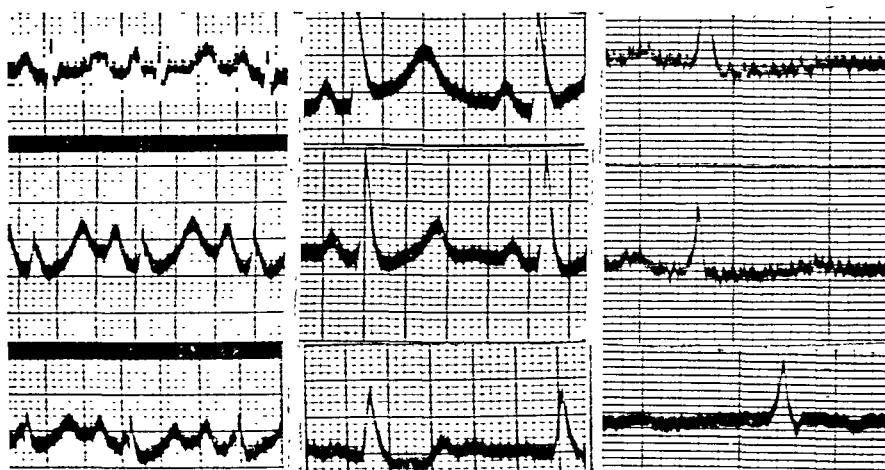

Fig. 5. Record taken 15 minutes after sympathectomy (right side). Operation complete. Wound sutured. Note changes similar to those of Fig. 4, perhaps due to persistent irritation of the cut nerve.

Fig. 6. Taken 40 minutes after operation. Patient conscious. Note return of *R* wave to normal. *T* waves still greater than normal.

Fig. 7. Two months after operation 20/7/33. Note return to form of electrocardiogram before operation. A similar record was obtained eight days after operation.

FURTHER OBSERVATIONS ON THE VASCULAR RESPONSES OF THE HUMAN LIMB TO BODY WARMING; EVIDENCE FOR SYMPATHETIC VASODILATOR NERVES IN THE NORMAL SUBJECT.*

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RECENT observations (4) have revealed differences between the vascular responses of the proximal and those of the distal parts of the human limb. A chief difference is that while body warming provokes a large increase of blood flow and skin temperature in hand and foot, it causes no more than a slight rise of forearm and leg blood flow and neither flushing nor warming of the skin of these parts, provided the circulation to the hand or foot is arrested. This difference between the parts is attributed to the distribution of arterio-venous anastomoses ; their presence in the extremities and their absence from forearm and leg.   It is not due to body warming releasing vasoconstrictor tone in the extremities and failing to do so more proximally, for while local nerve block flushes and warms the hand and foot these effects are wanting in the forearm and leg.

The body warming used was such as is employed to test the vasomotor reaction of the extremities, namely, covering the trunk with a blanket and immersing the legs or arms in hot water until the subject feels hot and sweats. It was noted, however, that different results are obtained if the warming is pushed to an unusual degree.   More rapid and intense heating, though provoking a relatively small further increase of bloodflow in the extremities, does cause a considerable increase in the forearm and leg together with flushing and warming of the skin.   It seems clear that some explanation other than release of cutaneous vasoconstrictor tone is required to account for these changes in the proximal part of the limb.   We now record further observations on this point ; they provide evidence in the normal subject for the existence of sympathetic vasodilator nerves to the skin.

Methods.

Strong body warming is accomplished by immersing two, or better three limbs in water maintained as hot as can be borne, 45 or 46°C., and by well covering all other parts of the body, leaving exposed only the face and the limb under observation. Immersion of two limbs only may fail to elicit the response.

Blood flow and skin temperature are measured as previously described (4); certain additional precautions are required for skin temperature, specially when the changes at different parts of the limb are compared. The limb should be supported not along its length but proximally and distally and should be well clear of any obstruction so that air may circulate freely around it. If the limb lies on a bench or pillow, the skin near the support may warm less than that on the upper surface presumably because some vessels supplying the skin are compressed by the weight of the limb.
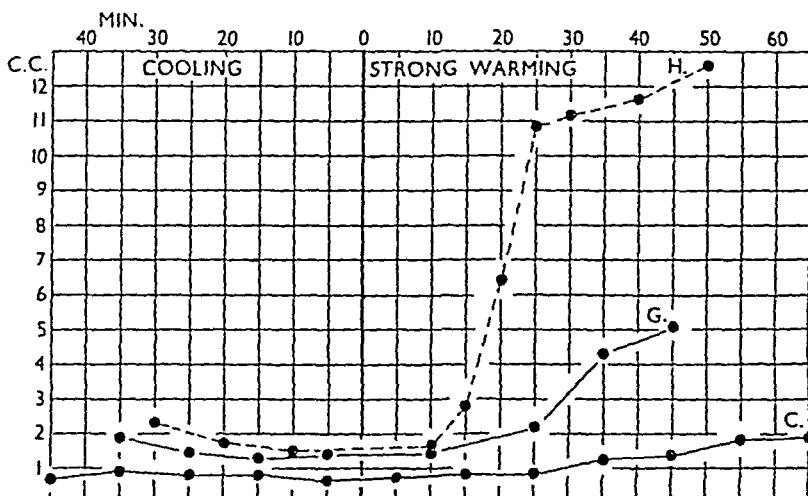

Fig. 1. Effect of strong body warming on forearm blood flow rate in three subjects, H., G. and C., aged 29, 45 and 51 respectively. In subject C. although heating was continued for an hour, forearm blood flow was hardly increased beyond that attained by gentler warming.

On the other hand, if the limb is raised only slightly from the bench the under part may warm more than the upper where heat is dissipated more freely. Since sweating is free, care must be taken that the adhesive plaster covering the thermal junctions does not become wet with sweat. It is our custom to smear both strapping and skin with vaseline. This prevents evaporation of sweat, and accentuates the rise of temperature; it does not obscure colour changes in the skin. Moreover, the sweat collects in beads beneath the vaseline; this provides a more certain test for recognising the presence or absence and degree of sweating than inspection and palpation of the unsmeared skin.* Most of the blood flow observations have been

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* By this method the normal " insensible perspiration " can be rendered visible.

made on the forearm. Blood flow rates, in the text and figures, are stated in c.c. of blood per minute per 100 c.c. limb volume enclosed within the plethysmograph.

Results.

General effects. Heated in this way, the subject soon becomes uncomfortably hot and body temperature rises rapidly, usually between 1 and 2°C. in a half to three-quarters of an hour. The respiration rate is increased slightly and pulse rate rises considerably, up to 130 beats per minute. As with gentler warming systolic blood pressure falls 5 to 10 mm. Hg. below the resting level but towards the end of heating may rise a little, no more than 5 mm. Hg.. Prolonged heating of this intensity is not well tolerated ; after about half an hour most subjects become restless and irritable ; a few have complained of precordial oppression and one has fainted. All complain of lassitude and fatigue and some of headache as after-effects.

Effect on limb circulation. In the fingers the blood flow rate may rise to between 30 and 40 c.c. and in the hand to between 20 and 25 c.c., the usual rates attained with gentler warming being 20 to 30 c.c. and 10 to 20 c.c., respectively.* As would be expected, there is but little increase in the temperature of these parts beyond that attained by gentler warming, namely, a few degrees below rectal temperature. In the forearm and leg vasodilatation begins usually 10 to 15 minutes later than in the extremities ; the blood flow rate increases to between 4 and 13 c.c., the skin becomes slightly flushed, sweats freely and its temperature rises. The flush is much less intense than that of reactive hyperæmia and is hardly so conspicuous as that of the flare. The appearance of the flush usually coincides with the outbreak of sweat, but we have seen the flush precede sweating and also profuse sweating without appreciable flushing.† There is no clear relation, even in the same subject on different occasions between the intensity of the flush and the degree of sweating. The examples given in Figs 1, 2 and 3 show that the vascular reaction varies considerably in different subjects. In some (Fig. 1, H and Fig. 2) it comes early, proceeds rapidly and is conspicuous, blood flow and skin temperature rising steeply in an S-shaped curve ; skin temperature may reach 34 or 35°C.. In others (Fig. 1, G and Fig. 3) the reaction is slighter and more gradual and may be long delayed. On occasion in some subjects it fails to develop even after heating for so long as an hour (Fig. 1, C). In general, the conspicuous effects are obtained from the younger subjects, though this is not always the case. It will be noted also that when heating is stopped (because of increasing discomfort to the subject), blood flow and skin temperatures have not usually reached steady levels but are still rising, though the rate of rise has declined. We shall not deal further with the

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* Because of the rapid filling of the veins of the hand at high rates of flow, these estimates are not altogether satisfactory and are approximate only.

† In a patient suffering from Raynaud's disease, the forearm sweated profusely for 20 minutes before the skin flushed and warmed ; the skin was smeared with vaseline.

vasodilatation in the extremities but pass to consider the mechanism of the response in the forearm and leg.

It is to be borne in mind that the intense heating causes a considerable rise of body temperature; this rise might possibly account directly for the small and gradual rise of skin temperature illustrated in Fig. 3. But it is clear that the more usual reaction (as in Figs. 2, 5 and 8), in which the rise of skin temperature is rapid and considerably exceeds that of the body, cannot be explained simply as a direct effect of rising blood temperature. This is amply confirmed by further analysis. To avoid confusing effects
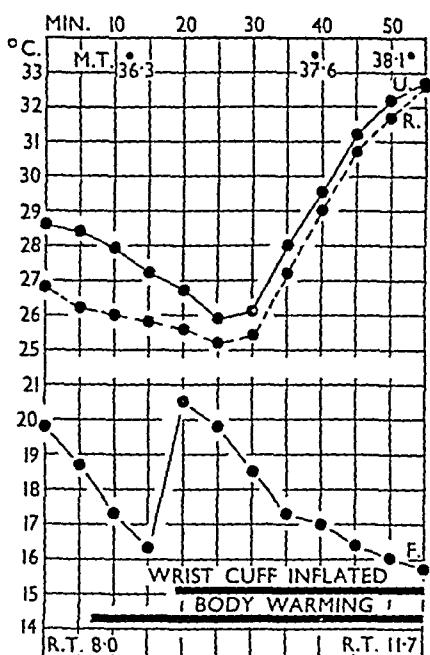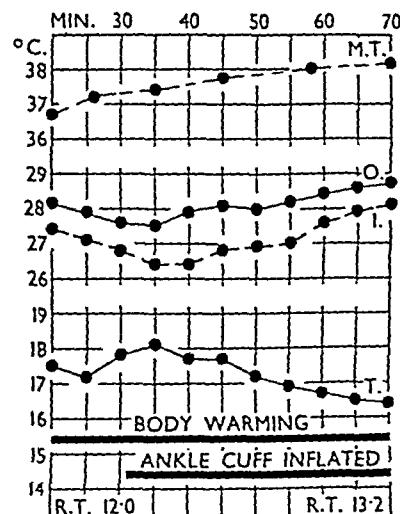

Fig. 2.   Fig. 3.

Fig. 2. Effect of strong body warming on skin temperature measured at middle of ulnar (U) and radial (R) sides of the forearm. F = index finger temperature. In this and subsequent figures M.T. and R.T. = mouth and room temperatures respectively; durations of body warming and inflation of cuffs indicated by heavy horizontal lines.

Fig. 3. Effect of strong body warming on skin temperature measured at inner (I) and outer (O) aspects of middle of leg; leg cooled for 20 minutes before chart begins. T = temperature of great toe.

from this factor, however, we have used chiefly subjects displaying conspicuous reactions and have ensured that the limb vessels are well constricted by sufficient rest and exposure to cool air before heating the body.

Influence of sympathetic ganglionectomy. Observations on patients before and after sympathetic ganglionectomy show that immediately after operation the circulation in forearm and leg is considerably increased; the

skin is flushed and hot, blood flow is raised.   The hyperæmia soon declines and after about a week the limb circulation returns almost to the pre-operative level.   Strongly warming the body then fails to provoke the vasodilatation observed before operation.   We recount these observations briefly.

One patient has been referred to in previous papers (3 and 4).   This man (S.K.) aged 31, suffered from thromboangiitis obliterans affecting the legs and feet mainly and also the hands. Forearm circulation was apparently normal.   Blocking the right internal cutaneous nerve above the elbow failed to cause flushing or warming of the skin on the anæsthetic ulnar side of the right forearm.   Strong body warming raised forearm skin temperature to 33°C. and blood flow rate to 5·2 c.c..   Under gas and oxygen anæsthesia right cervical ganglionectomy was performed for the relief of symptoms in the right hand.   The pleura was accidentally opened at operation and right pneumothorax resulted.   Except for low fever, 37·8°C., (100°F.) for two days after operation recovery was uneventful.   Immediately after operation the right arm was dry and hot while the left was moist and warm ; both were flushed.   The right palpebral fissure
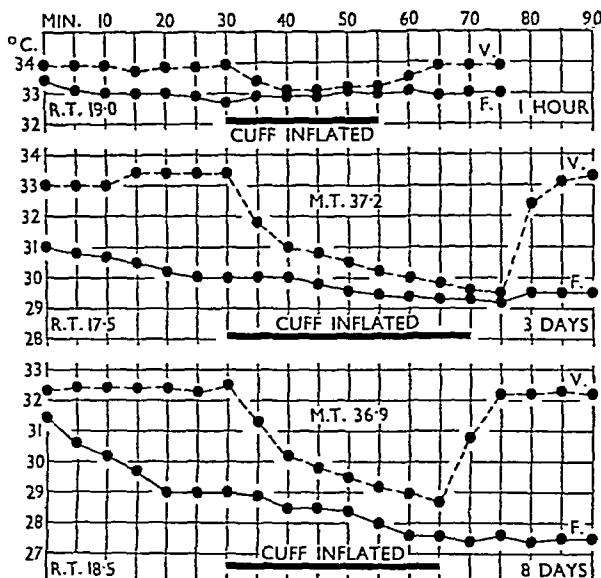

Fig. 4.   Temperature of dorsum of right forearm, (V over a vein and F remote from any obvious vein) 1 hour, 3 and 8 days after right cervical ganglionectomy.   Patient, S.K.

was narrowed, the eye sunken and the pupil contracted.   Charts of skin temperature after operation are shown in Fig. 4.   Temperature was measured throughout at the same two points on the dorsum of the right forearm, one (V) over a large vein draining the hand and the other (F) remote from any obvious vein, a little below the head of the radius, a region but little influenced in temperature by venous return from the hand.   On each occasion the forearm was exposed to room air and a sphygmomanometer cuff applied to the wrist.   After 30 minutes the cuff was inflated to arrest the circulation to the hand ; it was later released when the temperature effect of distal circulatory arrest was clearly displayed.   One hour after operation forearm temperature was high, and remained high when the venous return from the hand was arrested. The temperature at F remained steady at 33°C. throughout ; the temperature at V, initially only 1° higher, fell to that of F when the wrist cuff was inflated.   The resting forearm blood flow rate measured 19 hours after operation lay between 5·0 and 5·4 c.c..   On the third day the initial temperatures were lower.   That at F fell gradually on exposure to a level 1° lower ; it fell a little further when the cuff was inflated and rose a little when the cuff was released.   The temperature at V remained high so long as the circulation to the hand was free, but after arrest of this, fell at first rapidly and then more slowly to that of F.   Eight days after operation the initial temperatures were still lower ; the fall of F was greater and more rapid ; V behaved as on the previous occasion.

On the ninth day the blood flow rate was 1·2 c.c.. Thereafter temperature responses and blood flow remained unchanged until the patient left hospital well 20 days after operation. On the 10th and 11th days after operation strong body warming caused sweating, flushing and warming of the skin of the left but not of the right forearm ; it hardly altered right forearm blood flow, raising the rate only from 1·2 c.c. to 1·9 c.c.. The right forearm was now only doubtfully pinker than the left when that was cool, and paler than the left when that was flushed by body warming. The right hand, hot at first after operation, cooled a little during the next few days but was still warm and flushed when the patient left hospital.

In a second patient, M.A., a girl aged 23, right lumbar ganglionectomy was performed to relieve coldness and chilblains of the right leg and foot, paralysed by an attack of anterior poliomyelitis at the age of five. Observations were confined to skin temperature and the results agree with those of the preceding case. Temperatures were recorded from both great toes and from the upper part of both shins, an area little influenced by venous return from the foot. Before operation, the right leg was usually colder than the left. Fig. 5 shows that strong body warming
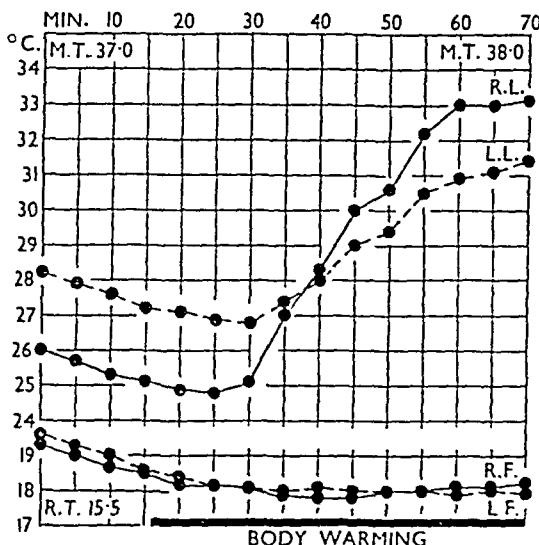

Fig. 5. Effect of strong body warming on skin temperature of right (R.L.) and left (L.L.) legs (middle of inner aspect), and of right (R.F.) and left (L.F.) great toes. Patient, M.A., before operation.

readily provoked flushing and warming of both legs. Note that in this instance the feet remained cold (but sweated) and it was unnecessary to inflate the cuffs applied to the ankles ; on other occasions the feet responded normally to body warming. After operation, the patient suffered from retention of urine for 48 hours followed by slight cystitis. She also developed a mild pneumonia of the left lung base. Her mouth temperature remained at 37·8 to 38·3°C. (100 to 101°F.) for about a week ; thereafter recovery was uneventful. Fig. 6 shows the temperatures of both legs and feet one day after operation and after they had been exposed to room air at 17·4°C. for 25 mins.. When first exposed both legs were equally flushed and hot to the touch. The left leg and foot cooled and paled normally on exposure ; the right leg and foot remained flushed and hot. The temperature of the right leg was uninfluenced by arresting the circulation to the foot ; it was lowered slightly and temporarily by the return of cold venous blood from the foot when the cuff was released. As in the previous case, the warmth of the right leg declined in the succeeding days ; the foot also became colder. Fig. 7 shows the temperature of both legs and feet exposed to room air 18 days after operation. Both legs were now about the same temperature, the right being slightly the warmer ; the right foot was about 4°C. warmer than the left ; legs and feet cooled on exposure. The same figure shows that while strong body warming provoked normal warming (also flushing and sweating) of the left leg, it had no effect on the right leg which continued to fall in temperature and remained pale and dry. Sweating was present on the front of the right thigh and knee ; the back of the thigh and knee remained dry.

Two other female patients were observed, one (B.J., aged 23) nine months after bilateral cervical ganglionectomy for Raynaud's disease, the other (M.M., aged 21) three years after bilateral lumbar ganglionectomy for acrocyanosis. In both, forearm and leg skin which failed to sweat when the body was heated, showed neither flushing nor warming.

These observations show that the vasodilatation provoked in the arm and leg by heating the body is dependent upon the integrity of the sympathetic nerves, as is already known for the hand and foot. The immediate effects of ganglionectomy also show that, just as in the hand and foot, vasodilatation in the forearm and leg can be brought about by interrupting these nerves.

Influence of local nerve block. It has already been noted that local block of cutaneous nerves to the forearm and leg fails to cause either distinct flushing or warming of the skin, although the skin becomes anæsthetic and
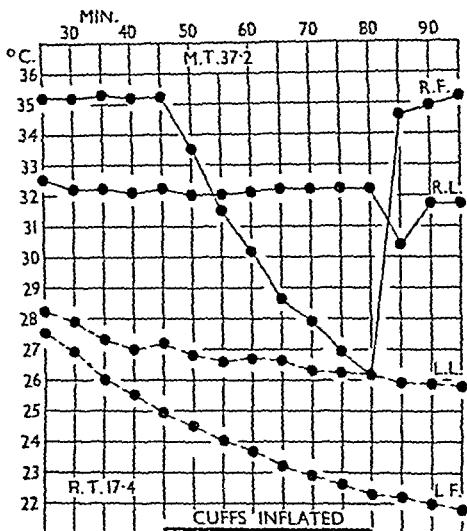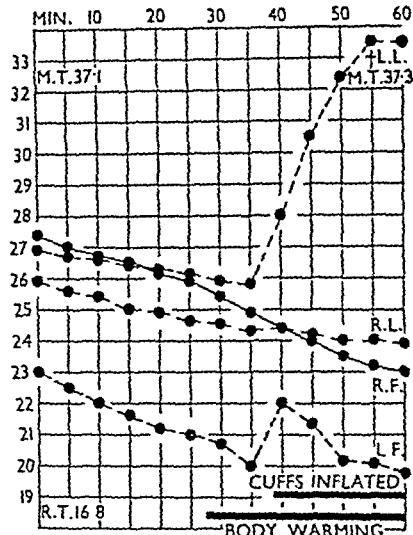

Fig. 6.

Fig. 7.

Fig. 6. Temperature of both legs (R.L. and L.L.) and great toes exposed to room air, one day after right lumbar ganglionectomy. Same patient as in Fig. 5.

Fig. 7. Effect of strong body warming on skin temperature of right (R.L.) and left (L.L.) legs (middle of inner aspect) and right (R.F.) and left (L.F.) great toes, 18 days after right lumbar ganglionectomy. Same patient as in Figs. 5 and 6.

smooth. That is to say, the removal of local constrictor tone does not materially increase circulation to the skin. But in view of the observations on newly sympathectomised limbs we thought that a cutaneous hyperæmia might be caused by removing vasoconstrictor tone not from skin but from the underlying muscle. It is known that increased blood flow through voluntary muscle readily raises the temperature of the overlying skin (3 and 4). Woollard and Phillips (7), however, find that local block of motor nerves, such as the dorsal interosseus and the motor nerves of the median of the forearm, gives no surface evidence of vasoconstrictor fibres being paralysed by the injection. We have made similar observations and our results agree with theirs. In two subjects the external popliteal nerve was blocked at the head of the fibula after cooling the leg by exposure to room

air ; in one 2% procaine and 1 in 50,000 adrenaline was used to paralyse the nerve ; in the other adrenaline was replaced by arrest of the circulation to the leg for a period of ten minutes. Although the anterior tibial muscles were completely paralysed in both instances the overlying skin neither flushed nor warmed. In one subject also the external popliteal nerve was blocked above the origin of the lateral cutaneous branch with procaine and adrenaline, thus producing an area of cutaneous anæsthesia over paralysed muscle in the front of the leg. The anæsthetic skin remained pale and cool. Thus, it seems clear that blocking the nerves to muscle, or skin, or both, in the forearm or leg causes no vasodilatation sufficient either to flush or warm the skin.

We are unable to account for this difference between the effects of blocking a mixed nerve to a part of the limb and those of section of the sympathetic nerves supplying almost the whole limb. It is possibly due to persisting tone in vessels proximal to those supplied by the blocked nerve. But other observations, with which we shall not deal now, suggest that nerve section has a dilator effect other than those due to the mechanical stimulation of cutting the fibres and simple removal of constrictor impulses. Trotter and Davis (6) suggest the liberation of irritative substances from degenerating nerves to account for the hyperæsthesia following nerve section. Dale and Richards (1) in dealing with the enhanced responses of vessels following section of their nerves speak of stimuli aroused by the processes of degeneration—(this view is briefly discussed in a previous paper (2)). We may note also that although both patients in whom the early effects of ganglionectomy were observed were febrile for a few days after operation, the limb vasodilatation can hardly be attributed to this. Present knowledge points to the existence of peripheral vasoconstriction during a febrile rise of temperature. The question remains for further investigation.

Other observations on local nerve block provide strong evidence that the flushing and warming of the skin provoked by body heating is brought about not by inhibition of constrictor tone but by stimulation of cutaneous nerves. Thus when the body is strongly warmed, normal skin over paralysed muscle flushes and warms as much as that over normal muscle ; on the other hand if the skin over normal muscle is rendered anæsthetic by nerve block or section and fails to sweat, then it remains pale and cool. If the nerve block is of short duration, as when arrest of the circulation is used instead of adrenaline, then as the anæsthesia passes off while body warming is maintained, the skin flushes and warms. Moreover, if the skin is first flushed and warmed and kept so by heating the body, and a cutaneous nerve is then blocked (the circulation to the hand being arrested), the area that becomes anæsthetic and ceases to sweat, pales and cools. In making these observations several points require attention which, if neglected, may lead to anomalous results. The area of anæsthesia should be large ; the temperature of a small anæsthetic patch may be influenced by that of the surrounding skin and specially by the venous drainage from skin situated distally. Secondly it is not always safe

to rely on anæsthesia alone as a sign that all sympathetic fibres to the area are blocked. For in some subjects although skin is rendered insensitive to stroking with a camel hair brush we have noted sweating and flushing appear in response to body warming, not only just inside the margin of but also in patches well within the anæsthetic area. The sweating is less than on
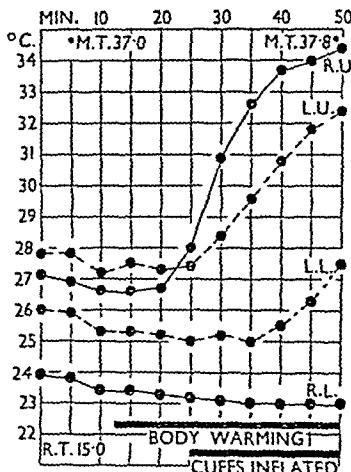

Fig. 8. Effect of strong body warming on skin temperature of the legs of a boy aged 17. His right external popliteal nerve had been torn through in a bicycle accident a year previously. It had been found impossible to suture the nerve at operation and the muscles of the front of the leg remained completely paralysed. There was also a large area of anæsthesia extending down the lateral aspect of the leg and on to the dorsum of the foot. The skin over the upper and anterior part of the leg was normally sensitive.

The legs were exposed at rest to room air at 15°C. and smeared with vaseline, sphygmomanometer cuffs being applied to the ankles but not inflated. Temperatures were recorded from points on normally sensitive (upper part of leg, R.U. in chart) and anæsthetic skin (lower part of leg, R.L.) overlying paralysed muscle in the right leg, from corresponding points on the left leg (L.U. and L.L.) and from both great toes. For simplicity, the toe temperatures are omitted from the chart. The legs being sufficiently cool, the body was strongly warmed, and when the toe temperatures began to rise both ankle cuffs were inflated to a pressure of 200 mm. Hg.. Normal skin over paralysed muscle (R.U.) warmed flushed and sweated quite as much as normal skin over healthy muscle (L.U.). The insensitive skin on the right leg remained pale, slightly cyanosed, dry and cold ; the skin on the corresponding part of the left leg flushed, sweated and warmed.

sensitive skin and would probably be unnoticed unless the surface were smeared with vaseline. The temperature of these patches may rise, though more slowly than that of normal skin. Again, in these subjects if the skin is first made to sweat and flush by body warming and the nerve is then blocked, sweating and flushing are not entirely abolished though they are reduced in these patches, the temperature of which does not fall.

The failure of flushing, sweating and warming to develop in an anæsthetic area is illustrated in Figs. 8 and 9. Fig. 8 shows the effect of body warming on the leg skin temperature in a boy whose right external popliteal nerve

had been accidentally severed above the origin of the lateral cutaneous branch a year previously. The muscles in the front of the leg were completely paralysed but the overlying skin was anæsthetic only over the lower part of the leg. The temperature of sensitive skin over the paralysed muscle in the right leg (R.U.) rose, even more steeply than that over normal muscle in the left leg. The insensitive skin in the right leg (R.L.) failed to warm, flush or sweat. Similar results were obtained in two normal subjects in
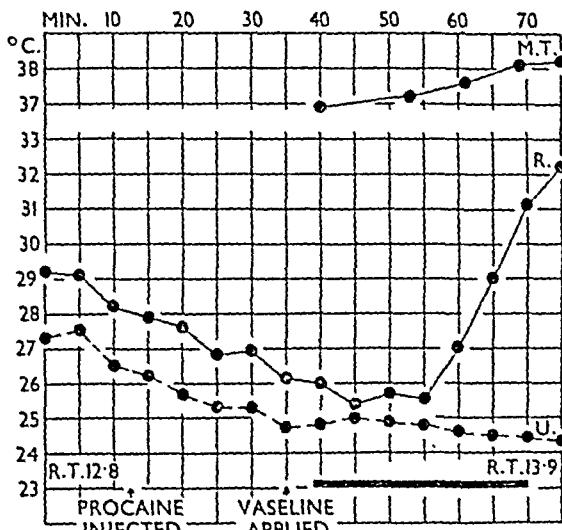

Fig. 9. Effect of strong body warming on skin temperature of ulnar and radial sides of right arm in a normal subject aged 23. The right arm was exposed to room air at 12·8°C., temperatures being recorded from radial and ulnar sides and from the index finger. A sphygmomanometer cuff was applied to the wrist. The trunk of the internal cutaneous nerve was blocked by procaine and adrenaline above the right elbow; 15 minutes later the skin over the ulnar side of the forearm was anæsthetic to stroking with a camel hair brush; the junction recording the ulnar temperature was situated well within the anæsthetic area. The forearm was smeared with vaseline and the body strongly warmed. When finger temperature (omitted from chart) began to rise, the wrist cuff was inflated. The radial skin flushed, warmed and sweated; the ulnar skin remained pale, dry and cool. Same subject as in Fig. 2.

whom this nerve was blocked by procaine and adrenaline. Fig. 9 shows the effect of blocking the internal cutaneous nerve of the forearm in the same subject whose normal reaction is illustrated in Fig. 2; the temperature of the radial side of the arm rises steeply when the body is warmed; that of the anæsthetic ulnar side continues to fall. Fig. 10 illustrates the subsidence of the vasodilatation on the ulnar side of the forearm as a result of blocking the internal cutaneous nerve.

The increased blood flow to the skin demonstrated by these observations must lead to warming and vasodilatation in the deeper tissues. It was previously shown (3 and 4) that, in the resting limb exposed to cool air, muscle as well as skin temperature gradually falls, muscle temperature

being several degrees higher. When the body is warmed the forearm being smeared with vaseline then skin temperature rises before and may for a time be higher than that of the underlying muscle. (For example, *see* Fig. 11).
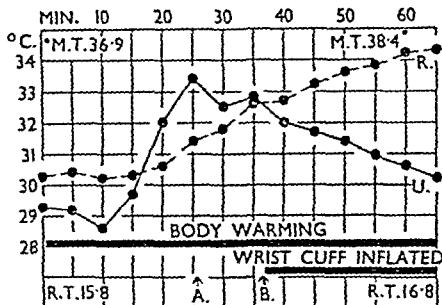

Fig. 10. Effect of blocking internal cutaneous nerve on forearm skin temperature while body is strongly warmed. Temperatures recorded from radial (R) and ulnar (U) sides of left forearm, and from index finger (omitted from chart). The body was strongly heated throughout, a sphygmomanometer cuff applied to the wrist and the forearm smeared with vaseline. After the forearm skin was flushed, sweating and warming, procaine and adrenaline was injected around the internal cutaneous nerve above the elbow (at A). Anæsthesia developed rapidly and after 10 minutes involved the whole ulnar side of the forearm. The vaseline and underlying sweat was gently wiped off; then vaseline was replaced and the wrist cuff was inflated to prevent warming of the forearm by venous return from the hand (at B). The anæsthetic skin ceased to sweat, paled and cooled. A control observation carried out in the same way with injection of procaine and adrenaline but without producing anæsthesia showed that ulnar skin continued to rise in temperature.
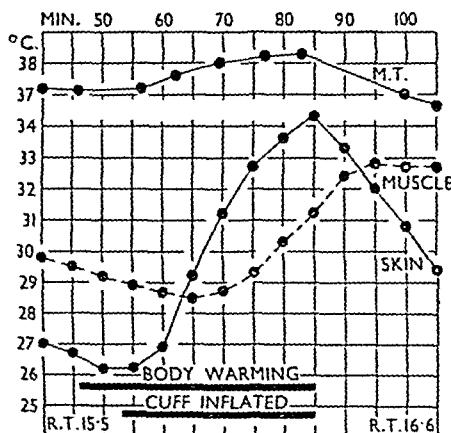
Fig. 11. Effect of body warming on temperature of skin and underlying muscle. Temperatures recorded from skin on dorsum of forearm and from underlying muscle (a copper constantin thermal junction was inserted for a length of 4·5 cm. into the arm, the tip lying about 2 cm. beneath the surface). Temperatures also recorded from index finger (omitted from chart). When the body was warmed skin temperature rose steeply before and to a higher level than that of the muscle. At 85 minutes heating was stopped and the blankets removed. Skin temperature fell steeply; that of the muscle continued to rise for some time.

Comment.

These observations supplement the earlier ones by Lewis and Pickering
(5) on patients suffering from Raynaud's disease. In such patients exposed
to a suitable temperature ulnar anæsthesia with its inhibition of vaso-
constrictor tone fails to release a vascular spasm in the hand, but under the
same conditions warming the body relaxes the vessels. The last effect occurs
through sympathetic paths, but is not due to simple inhibition of vaso-
constrictor tone, being prevented by anæsthetisation of the ulnar nerve,
and must be attributed to active vasodilatation. Lewis and Pickering (5)
conclude that the vessels in the human arm are supplied with vasodilator
sympathetic fibres. They failed, however, to obtain evidence of these in the
normal hand ; as they point out, the loss of vasoconstrictor tone so raises
the temperature of this part that the accession of vasodilator impulses has
little chance of displaying itself in a further rise of temperature. We have
seen that in the normal forearm and leg, the loss of constrictor tone produces
so little increase in the circulation that the effects of vasodilator stimulation
are readily displayed. It is to be remembered that the cutaneous vaso-
dilatation is associated with sweating. The available evidence is insufficient
to decide definitely whether the dilatation is caused through nerves supplied
to the vessels themselves, though this seems likely, or whether it is the result
of activity of the sweat glands which are also stimulated through sympathetic
paths.

A word remains to be said about these observations and those previously
recorded (4) from a more general point of view. We interpret them as
indicating two means of defence against a rise of body temperature. The
first is brought into action by relatively gentle heating and consists chiefly
of a dilatation of the arteriovenous anastomoses in the extremities, caused
by the inhibition of vasoconstrictor tone. The blood is cooled as it returns
through the capacious superficial venous plexus of the limbs. The second is
added when the heating is more intense and consists mainly of a general
dilatation of the cutaneous vessels associated with sweating, both of which
are caused by the stimulation of the sympathetic nerves, and serve to
increase the dissipation of heat from the skin.

Summary.

1. Further observations on body warming in man show that when this
is pushed to an unusual degree a considerable vasodilatation develops in the
proximal parts of the limbs.

2. The vasodilatation is brought about by stimulation of cutaneous
sympathetic nerves.

3. These and previous observations are interpreted as indicating two means of defence against a rise of body temperature.

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THE PATHOLOGICAL CHANGES IN THE ARTERIES SUPPLYING THE FINGERS IN WARM-HANDED PEOPLE AND IN CASES OF SO-CALLED RAYNAUD'S DISEASE.*

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Material.

BEFORE attempting to interpret the changes seen in sections of the vessels of the hand in cases of what is commonly called " Raynaud's disease," it seemed desirable thoroughly to examine the corresponding vessels from a series of subjects who during life presented no Raynaud phenomenon. To obtain this material I enlisted the help of Dr. B. B. Gelfand of New York, then working in my laboratory, and I here gratefully express my indebtedness to him for his careful and patient assistance. The plan adopted was to examine a large number of people in the wards of the hospital who, while sufficiently well to give a willing and clear account of their past, were known to be suffering from incurable diseases. The history and diagnosis of the serious malady being already available, special enquiries were directed to the past and present state of the hands. The patient was regularly asked whether he had been a warm or a cold-handed person, if friends had remarked on the feel of the hands, if chilblains had been experienced, if the hands had ever become unusually discoloured in cold weather or after bathing in cold water, and other relevant questions. The replies were checked by repeated observations on the hands exposed to the air of the ward in cool or cold weather. The hands were closely examined not only to gain a general idea of their temperature and colour, but they were searched for nutritional changes in the skin and the pulses were felt. In nearly all cases one hand, or both, was soaked for 3 minutes, or more if necessary, in water at 46° to induce full dilatation of the vessels ; the fingers were then examined for palpable pulsation, and the pads of the fingers for capillary pulsation ; the 2nd and 4th finger of the hand were always looked at first and with particular

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TABLE I.

Age and Sex.	Occupation.	Death from	B.P.	Hands.	Heated fingers.		Digital arteries.		
					Cap. pulse.	Palpable pulse.	Intimal thickening.	Medial sclerosis.	
1. M. 21	Motor mechanic	Leukæmia	84/50	Warm			Slight or none	None	
2. F. 24	Housework	Pulmonary tuberculosis	98/52	Warm	+	+	Slight or none	Slight or none	*See* Fig. 1.
3. F. 25	Housewife	Mitral stenosis. Heart failure	115	Cold	+	+	Slight to moderate general	Slight or none	Chilblains since childhood.
4. M. 28	Chauffeur	Hodgkin's disease	116/78	Warm	+	+	Slight general to moderate patchy	Slight	Chilblains, single attack.
5. M. 39	Wood-worker	Carcinoma of lung	104/70	Warm	+	+	Slight patchy	Slight	*See* Fig. 2.
6. M. 42	Labourer	Carcinoma of lung	124/90	Warm	+	+	*Slight general or patchy*	Slight	
7. M. 43	Tyre fitter	Carcinoma of bronchus	150/100	Warm	+	+	Slight general	Slight	
8. M. 45	Caretaker	Leukæmia	122/80	Warm	+	+	Slight general or patchy	Slight	
9. M. 50	Signalman	Carcinoma of bronchus	104/70	Warm			Slight to moderate general	Slight	*See* Fig. 5.

10. M. 52	Soldier and cook	Mediastinal new growth	122/72	Warm	+	+	Slight general occasionally moderate	Slight	
11. M. 53	Porter	Aortic aneurysm	138/80	Warm	+	+	Slight general occasionally moderate	Slight	Radial and ulnar artery showed mod. intimal thickening, solid arterioles of fingers.
12. F. 57	Waitress	Syphilitic aortitis	128/40	Cold	+	+	Slight general or patchy, rarely mod.	Slight to moderate	Slight intimal thickening in digital arterioles.
13. M. 59	Stage hand	Carcinoma of lung	104/62	Warm	+	+	Slight general or occasionally moderate	Slight	Radial and ulnar artery showed moderate intimal thickening.
14. M. 64	Attendant	Carcinoma of œsoph.	84/48	Warm			Slight to mod. Occasionally considerable	Moderate	
15. M. 66	Painter	Carcinoma of neck	170/88	Warm	+	+	Mod. general, occasionally considerable	Moderate	Heavy sclerosis in media of larger arteries. Intimal thickening of digital arterioles. See Fig. 15.
16. M. 73	Baker	Abdominal carcinoma	144/78	Warm	+	+	Moderate general	Slight to moderate	Intimal thickening of digital arterioles.
17. M. 73	—	Intestinal obstruction	120/72	Warm	+	+	Slight to mod. general or patchy	Slight to moderate	Intimal thickening of digital arterioles.
18. M. 76	Labourer	Rectal carcinoma	118/78	Warm	0	+	Moderate general to considerable	Slight to moderate	Intimal thickening of digital arterioles.

Portions of the radial and ulnar arteries, the whole palmar arch, and the whole length of the two digital arteries from both the 2nd and 4th right finger were removed for examination.

The medial coats of none of these vessels presented more than slight sclerosis. In the palmar arch and in the digital arteries of both fingers there was slight intimal thickening of a patchy kind. The thickening was an intimal hyperplasia, the tissue being pervaded by fine wavy elastic fibres, interlacing but in general parallel to the elastic lamella (Fig. 10). In one part of the palmar arch only was the intimal thickening a little greater. The arteries of the 2nd finger were both cut serially in their whole length, that is to say from their origin in the palmar arch onwards. The arteries were unusually large and unobstructed throughout; the intimal coat presented less change than is commonly found in warm-handed people of the same age.

The small arterioles of the fingers were normal.

Case 2. *Raynaud's disease (mild form).* B.B., a telegraphist of 58 years was admitted to hospital suffering from advanced carcinoma of the breast with metastases in February, 1935. She died in April shortly after leaving hospital.

While in hospital she said that her hands had been cold, winter and summer, as long as she could remember. Chilblains had not been experienced. For about 28 years she had noticed while bathing in the sea that all the fingers of both hands would become white and numb, starting at the tips and gradually extending to the bases. The circulation could be restored by vigorous rubbing. She stated that she remembered no attacks induced by exposure to cold air.

On examination the skin of the fingers seemed quite normal in colour and texture. After heating the right hand in water at 46° for 3 min., capillary pulsation was displayed in all the finger tips and in the palm, and pulsation could be felt at the bases of all the fingers. Blood pressure was normal.

Post-mortem. After death dissection was limited to the right hand. The radial and ulnar arteries, the palmar arch, and the whole length of both arteries from the 2nd and 4th right fingers were removed for examination. On section none of these vessels showed more than slight sclerosis of the medial coat; none presented more than slight intimal thickening and this usually only from place to place. The two digital arteries from the 4th finger were cut serially in their whole lengths. Fig. 3 represents the least and Fig. 4 the greatest change found in these arteries. In no artery were the changes as great as those often found in warm-handed people of the same age.

The small arterioles in these fingers showed slight intimal thickening.

Case 3. *Raynaud's disease (moderate severity).* W.A.S. was reported in a previous paper as Case 8. A thin unemotional man of 44 years attended the out-patient department in 1926 and onwards for mitral stenosis and auricular fibrillation. He was under observation for four years. He had

suffered from chilblains of the feet and also from attacks of white or blue fingers in cold weather for many years. One or more fingers, occasionally four fingers on one hand, would become blanched, cold and numb ; the pallor would last minutes or hours, be replaced by blueness and the hands would finally become red. Sometimes the fingers would become blue from the start. The finger tips were always first affected, the blanching or blueness spreading up to their roots. The thumbs were not affected. They recovered in response to local warmth. The attacks were frequent in cold weather, rare in summer, but occurred occasionally even on warm days. Excitement according to the man seemed sometimes to cause attacks. Pain was not experienced, no complaint of the feet or face was made, and there was neither history nor sign of loss of substance in the fingers. Visual disturbances, and blood in the urine, had not occurred.

The hands between the attacks were high coloured, often mottled with red and various cyanotic colours (from XI to XIV). They were usually cold, but occasionally quite warm. Single fingers were seen in a blanched state or of a pale cyanotic colour on a number of occasions. Attacks were induced by immersion of a hand in water at 15°. Fingers 4 and 5 of the right hand were those most usually affected. The colour-temperature curves of these fingers were those of the fingers of people without symptoms.

The condition of his fingers remained unaltered during the 4 years. In March, 1930, he acquired bronchitis. He developed venous congestion and infarction of the lungs in June, 1930, and died within three days.

Post-mortem. Severe stenosis of the mitral valve, extensive ante-mortem thrombosis of the left auricle, infarction of the lungs, congestion of veins and liver, and an ulcer of the duodenum were found after death.

A dissection was made of his right hand and the whole palmar arch and all the digital arteries of fingers 2, 3, 4 and 5 were removed. Serial sections were cut through the palmar arch, through the greater part of the length of a single digital artery of the 2nd and 5th fingers, and through the whole length of both arteries of the 4th finger (his worst finger). These arteries all showed very similar changes, namely, some sclerosis of the media and moderate and uniform intimal thickening (Fig. 6). The elastic lamella was well defined, though sometimes broken. The intima was hyperplastic ; in its outer layer there were a few wavy coarse elastic fibres running parallel to the lamella and in its inner layers finer elastic fibres arranged more longitudinally ; whether stained for elastic tissue (Fig. 6) or by hæmatoxylin or eosin, the intimal tissue presented a very uniform appearance when traced around the circumference of the vessel. The lumen, though often small owing to the contracted state of the vessels, was nowhere nearly obliterated. There was no sign of recent inflammation or of ante-mortem clot in these digital arteries.

One stretch of the palmar artery presented more conspicuous and more irregular intimal thickening and a broken elastic lamella, and contained more connective tissue (Fig. 7).

No essential difference could be found between the changes in these digital arteries and those found among warm-handed people of a little greater age, and described in them as moderate general intimal thickening; this statement applies not only to the degree but to the kind of intimal thickening.

Summary. Clinically this case of Raynaud's disease was one of moderate severity without evident signs of vascular disease and without nutritional changes, and unprogressive. The digital arteries all exhibited a moderate general intimal hypertrophy of very uniform appearance and thickness, but no greater in degree than is found in some elderly people whose hands are ordinarily warm. The lumens of the arteries were narrowed but nowhere obstructed.

Case 4.   Raynaud's disease with small healed necroses.   A.Z., a married woman of 33 years was admitted to hospital on April the 18th, 1935, suffering from salpingitis developing into septicæmia, which killed her on May the 5th, 1935.

During the early days of her admission she gave the following history. She had suffered from cold hands and feet, winter and summer, as long as she could remember. For 10 years, chilblains of fingers, toes and backs of heels had occurred annually. For 14 months, including two winters, she had noticed attacks of blanching or purple discoloration of all five fingers of both hands ; the discoloration would begin at the tips and spread to the bases of all fingers, and they would become numb. White fingers were seen only when dressing on cold mornings. Immersion of the hand in cold water or exposure out of doors on cold days always made her fingers dark blue in colour. Rubbing the fingers or immersing them in hot water would restore the colour and the fingers would then tingle.

Her mother, brothers and sisters had all suffered from cold hands and feet and chilblains, but not from attacks of discoloured fingers. The patient had not experienced hæmaturia. Her blood pressure was 122 syst. and 80 diast..

On examination the skin of the hands showed scaly eruption, part of a generalised icthyosis. The skin of all the fingers was quite mobile, but flexion of the fingers was a little limited. The 3rd right finger and the 2nd and 5th fingers on the left side had lost a little substance at their tips, as shown by scars. After soaking in hot water the skin of all the fingers was red, and slight capillary pulsation was seen in all fingers, but not at their tips. Weak pulsation was felt in some of the digital arteries. Her radial pulses were normal.

Post-mortem.   Both Fallopian tubes contained pus, so did the inflamed cervix uteri. Pus was present in both knee joints. The main arteries presented little atheroma and the veins were patent and healthy. Pieces of the radial and ulnar arteries were excised from the right hand ; the palmar arch, and both digital arteries from each of fingers 2, 3 and 4 were dissected out in their whole lengths.

All these arteries when examined histologically showed a slight or moderate sclerosis of the media. The intima of the radial artery was normal, that of the ulnar showed slight general thickening, and that of the palmar arch was in the same state.

Arteries of the 2nd right finger. Both these vessels presented a moderate grade of general intimal hyperplasia. Examined in serial sections, the lumen of one of these arteries was very much narrowed throughout, though the vessel was patent. The other was replaced by several small arteries, themselves presenting gross intimal thickening with small or actually obstructed lumina.

Arteries of the 3rd right, scarred, finger. These were examined in serial sections from end to end. The digital artery on the radial side presented advanced intimal thickening throughout its course. It is illustrated after it has branched in Fig. 11. The well defined elastic lamella was broken here and there. The outer layers of thickened intima contained elastic fibres, the inner layers were formed by a cellular substance with no elastic tissue and little collagen (*vide infra*). The lumen of this artery when present was always very small ; the artery was completely blocked in several places by organising clot ; and in two places near its base the cellular intimal thickening had completely occluded the vessel. The digital artery on the ulnar side presented a moderate and fairly uniform intimal hyperplasia (Fig. 16) throughout its course ; the lumen contained normal red blood cells, except at two places where it was obstructed by quite recent blood clot.

Arteries of the 4th right finger. These were also examined in serial sections from end to end. At their bases the lumens of both arteries were rather small owing to the usual intimal hyperplasia. Traced into the finger this hyperplasia gave place to a much greater intimal thickening of distinct type. Both arteries were similarly affected. Loosely arranged tissue consisting largely of big well-nucleated cells with vacuolated protoplasm closed or almost closed the lumen ; here and there the much narrowed lumen was finally closed by recent clot, in one place hæmorrhage had apparently occurred into the new tissue. Remains of the hyperplastic intima in the inner layers sometimes gave an appearance resembling an artery within an artery. The last appearance was usual in the arterioles of this and other fingers ; Professor Turnbull regarded the vacuolated cells (in the arteries of this and of the 3rd finger) as lipoid macrophages or " foam cells " and called attention to the resemblance between the intimal thickening and that often found in the arterioles of " malignant hypertension." No thrombi were found in the veins.

Summary. Clinically the case was one of severe Raynaud's disease with scarring of finger tips from healed necrosis and signs of digital arterial obstruction, but of only 14 months duration. The digital arteries were much diseased. Moderate hyperplasia of the intima was present in many places.

In others the intimal thickening was of distinct cellular type and the lumen greatly reduced or lost. Here and there the lumens were finally blocked by recent or organising thrombus. The changes were very similar to scarred and unscarred fingers.

Case 5. Raynaud's disease with small healed necroses. F.C., a married woman, was first seen in her 60th year in October, 1935, for Raynaud's disease. In July, 1936, a carcinoma of the stomach was diagnosed ; she refused early operation and died of obstruction of the splenic flexure in March, 1937, pulmonary embolism following an exploratory operation.

She stated in October, 1935, that she had been treated for myxoedema for 27 years and that she had suffered from attacks of white or blue numb fingers for 3 years, on exposure on cold days, winter or summer, or when washing in cold water. Her fingers were swollen from time to time and the index fingers had developed little painful sores. Her feet were often cold, numb and painful. There had been no chilblains. She had flat feet.

When she came for examination her hands presented a characteristic attack, fingers 2 to 5 on both being livid (tint XV) to their roots and cold. The hands were puffy, the backs pitting distinctly with pressure. The skin of the fingers was mobile but the tips of all the fingers, with the exception of the 1st and 5th fingers on the left side showed small scars, with distinct loss of pulp in most of these fingers (including R2 and R 4, L 2 and L 4). Capillary pulsation was not found in the finger tips after heating the hand. Distinct pitting to pressure was present over the forearms and shins, and telangiectases were present on the cheeks ; she stated that she had noticed puffiness of the eyes sometimes ; but the skin of all these parts was normally mobile.

Post-mortem. A large ulcerating carcinoma was found in the stomach, infiltrating the splenic colon and obstructing it. The aorta showed a little atheroma, the coronary and iliac arteries were atheromatous but unobstructed. The right femoral vein was thrombosed and an embolus blocked the right pulmonary artery. Pieces of skin from forearm, hand and fingers were examined for evidence of early scleroderma, but none was found. Pieces of the radial and ulnar arteries were excised from the right hand. The palmar arch, and both digital arteries of each of fingers 2 and 4 were dissected out in their whole length. These vessels were all examined. They presented little medial sclerosis. The radial artery showed a little generalised intimal thickening.

The right ulnar artery and the palmar arch were plugged by connective tissue similar to that found in the digital arteries.

Arteries of right finger 2. On the radial side of this finger distally, the only artery found is shown in section in Fig. 9. It consisted of a very wide and vascular adventitia, a thin circle of medial muscle cells, also vascularised, and a central core of dense fibrous tissue to one side of which the clearly recognisable elastic lamella was displaced as a crumpled mass. In the

middle and at the base of this finger, the digital artery of the radial side was plugged by dense, poorly nucleated almost homogenous connective tissue, containing fine elastic fibres in its outer layers. Its media and the central core of connective tissue were both vascularised. The artery on the ulnar side was similarly plugged in its length. In both plugged arteries the elastic lamella was well defined and intact.

Arteries of right finger 4. The lumens of these two arteries were plugged in their length by substance (Fig. 8) having just the same appearances as have been described in the vessels of the 2nd finger. The middle stretch of the artery on the ulnar side was disorganised in much the same way as that depicted in Fig. 9. No thrombi were found in veins of these fingers. Small arterioles in the sections usually showed little change.

Arteries of left hand. These were similarly dissected and examined. The left ulnar artery was completely, and the palmar arch partly, filled with connective tissue containing occasional new vessels. The connective tissue in the palmar arch contained much scattered hæmosiderin. The media of both arteries was vascularised.

Arteries of left fingers 2 *and* 4. The four digital arteries of these fingers were all closed by fibrous plugs, containing scattered masses of hæmosiderin. The media was in all instances vascularised.

Summary. Clinically this case, a woman of 60 years, was one of severe Raynaud's disease of 5 year's duration, displaying evidence of arterial disease and of nutritional changes in the fingers. Histologically the digital arteries presented no clear evidence of intimal hyperplasia, though such tissue may have become incorporated in the outer layers of the arterial plugs ; even so the walls of the arteries could not be held to exhibit as much basic change as is to be found in those of most warm-handed people of the same age. The obvious abnormality was the plugging of the arteries by dense connective tissue, clearly consisting of organised thrombi. The uniform appearance of these plugs in the arteries (the only exception being an occasional disorganised vessel such as is shown in Fig. 9) suggested that they were all of more or less similar age, and they were presumably formed early in the illness ; the presence of hæmosiderin in the plugs of the left, and its absence from the right, hand vessels may signify some differences in the age of the clots on the two sides.

Case 6. *Raynaud's disease with ulceration.* J.N., a married man of 47 years, was admitted in August, 1936, for the condition of his hands. For 20 years he had developed sore places on his finger tips from time to time, and especially during the winter. Attacks of blueness of the fingers had been noticed in cold weather over the same period but not before the sore places first appeared. The sore places gave rise to much burning pain. All the fingers with the exception of the left thumb had been ulcerated. He made no complaint of his feet.

TABLE 2.

Six cases of Raynaud's disease.

Case, sex, age.	Cold hands	Attacks for	Frequence	Provoked by	Nutritional changes in fingers.	Capillary pulse.	Lesions.
1. F. 53	Always and chilblains	24 y.	Weekly	Cold air*	None	Yes	Palmar arch and digital arteries show less change than in warm-handed people of same age.
2. F. 58	Always. No chilblains	28 y.	Infrequent	Sea bathing only	None	Yes	Palmar arch and digital arteries show less change than in warm-handed people of same age.
3. M. 44	Always. Chilblains	20 y.	Frequent in winter	Cold air or water. Excitement	None	Yes	Palmar arch and digital arteries show moderate general intimal hyperplasia; more than in warm-handed people of same age.
4. F. 33	Always. Chilblains	14 m.	Frequent	Cold air or water	Scarred at tips	No	Digital arteries show moderate to advanced intimal thickening, the lumens being greatly reduced in places or blocked by cellular tissue, or occasionally by recent or organising thrombi. Similar changes in scarred and unscarred fingers.
5. F. 60	Always. No chilblains	5 y.	Frequent	Cold air or water	Scarred at tips	No	Digital arteries all plugged by organised thrombi, all of similar appearance, occasionally recanalised; palmar arteries similarly plugged.
6. M. 47	—	20 y.	Frequent	Cold air	Scars and ulcers at tips (20 y.)	No	Digital arteries show limited intimal hyperplasia and lumens plugged by organised, sometimes recanalised, thrombus. Amputation finger.

* A daughter also affected.

When examined the heart and large arteries showed no abnormality clinically. The terminal phalanges of all the fingers, including the thumbs, were cold and of deep blue colour. Scars were found at the tips of all the fingers except the left thumb, the only one which had never been ulcerated. The terminal sections of the 3rd, 4th and 5th left and of the 3rd and 5th right fingers were deformed and shrunken ; the 4th right finger was of full length, swollen and bulbous at its end. The tip of the 3rd left finger was ulcerated. The X-ray showed partial absorption of the terminal phalanges of the 3rd and 5th left and 3rd and 5th right fingers, with destructive changes in the corresponding terminal joints. A scar was found on the edge of the upper pole of each pinna.

When the feet were kept in hot water for 15 min. the fingers became warm, with the exception of the 3rd, 4th and 5th of the left hand. After cutting off the blood flow to the warm hand, reactive hyperæmia was delayed in the 3rd finger and in that only. When heated this finger failed to show capillary pulsation.

The 3rd left finger failed to heal under treatment and continuing to be very painful was amputated 2 months after admission.

Biopsy. The vessels of the amputated 3rd left finger were first examined opposite the middle phalanx. One digital artery appeared to be normal. The other was obliterated by almost homogenous connective tissue, sparsely nucleated, and recanalised (Fig. 12) ; the vessel wall was little changed except that it participated in new growth of blood vessels, and was therefore unusually vascular. Near the base of the finger both main arteries were plugged by the same sparsely nucleated almost homogenous connective tissue containing fine elastic fibres ; one of these plugs was slightly recanalised; the elastic lamella of these arteries was broken here and there. In some parts of their courses where the lumen of the artery was still patent, the intima was evidently hyperplastic. No hæmosiderin was seen in the plugged vessels. After examining the whole circumference of the base of this finger in section, it was clear that its circulation was carried only by quite small arteries or arterioles, the intimal layers of the former being often much thickened. The veins showed no recent thrombosis.

Summary. This case, one of severe Raynaud's disease, was peculiar in the occurrence of sore places on the finger tips over so long a period as 20 years, and in the emphatic statement that attacks of discoloration did not precede the sore places. Histological examination of an amputated ulcerated finger revealed digital arteries, the coats of which were affected only a little by disease, exhibiting only occasionally a limited hypertrophy of the intima. The lesions responsible for vascular obstruction, at the base of the finger and elsewhere, were old recanalised thrombi ; and the circulation to the finger apparently had been carried on precariously through these channels and through small anastomotic arteries, themselves presenting intimal hypertrophy.

General note on the adventitia and media.

Comparing the outer coats of the digital arteries in these cases of " Raynaud's disease " with those from warm-handed people, I have been unable to find any consistent difference. In particular the digital arteries in my cases of Raynaud's malady display no more than the strong musculature of the media that is usual in these vessels.

Discussion.

The six preceding case reports are summed up together in Table 2. Pickering and I divided cases of so-called " Raynaud's disease " into groups of which the first was :—

Intermittent spasm of digital arteries. Cases 1, 2 and 3 of Table 2 belong to this group ; and so far as I can ascertain these are the only cases of the kind in which the digital arteries have as yet been examined. Cases 1 and 2, the first relating a family predisposition, were both very mild cases, the attacks of discoloured fingers being provoked infrequently over a period of very many years. In these two women of 53 and 58 years, respectively, the arteries showed less change than is to be found in many warm-handed people of the same ages. Case 3 was a case of greater severity, attacks being frequent in winter and induced by immersing the hands in cold water. The intimal hyperplasia found was moderate in degree ; more than has been found in warm-handed people of the same age, though no more than that seen in older subjects whose hands are warm.

The findings in the first two subjects make it clear that cases occur in which no structural change exists that can be regarded as contributing to the attacks of transient discoloration of the fingers, which the cases display. Of the third case the same may be said, since it is clear that a moderate or even considerable amount of arterial disease can occur without provoking attacks (*see* subjects 15 and 18 of Table 1). To explain the attacks in cases of the type illustrated by these three, it is necessary to assume overaction of the musculature of the arteries concerned.

Intermittent spasm with local nutritional changes. Cases 4 and 5 of Table 2 belong to this group. I consider these two cases together because they both presented scarred finger tips, the result of tiny necroses, and because the skin of the finger was quite free from necrosis at the time of death. There appear to be no reports of similar cases to add to these. In Case 4 the intimal lesions included moderate hyperplasia, a distinct type of cellular thickening, greatly narrowing or occluding the lumen, and fresh or organising thrombi. The lesions were similar in scarred and unscarred fingers. In Case 5 the intimal hyperplasia was less advanced than is found in warm-handed people of the same age. All the digital arteries examined were plugged by organised clot, arteries of scarred and unscarred fingers being equally affected. In these examples the digital arteries were so damaged

as conduits by disease that little blood supply could have passed through them to the tissues. It is to be noted that in both cases the history was of symptoms for but a few winters, with which the obstructing lesions were in the main consistent. It is suggested that in one of these cases (Case 5), the malady began in simultaneous thromboses of a number of digital vessels.

Case 6 of this Table 2 I consider together with the first four cases of Table 3, derived from other sources. These cases all presented attacks of discoloration during life, but with one exception were of but a few years duration. I group them together because they all showed relatively massive unhealed necrosis of the fingers at the times the specimens were obtained. The feature of these cases, in which moderate intimal hyperplasia was sometimes distinguishable, was thrombotic obstruction of the arteries, seen in various stages of organisation. In one case of this group (Case 4 of Table 3), the lesions found were similar in the actually necrosed or merely discoloured fingers.

Bilateral gangrene of digits. Cases 5 to 8 of Table 3, collected from past records, are grouped together for separate treatment because they gave no history of spasmodic attacks, but only of an isolated attack of persistent discoloration quickly culminating in relatively massive necrosis of fingers, a history in itself suggesting thrombosis as the primary cause of mischief. The lesions found were similar to those of the last group, the predominant lesion being occluding thrombus in various stages of organisation. In one case of this group too, (Case 8 of Table 3) the lesions found were similar in the actually necrosed and in the merely discoloured fingers, an observation which shows that the thrombosis was not secondary to the necrosis, but is consistent with its being responsible for necrosis. The resemblance between the morbid anatomy in this group and the last, and the usual short duration of the symptoms in both groups, leads us to ask if cases included in the two groups are not for the most part pathogenetically alike. Thrombi occluding both arteries to a finger may lead at once to persistent discoloration terminating in necrosis. In another instance, where such occlusions hindered bloodflow to a less extent, the obstruction might proclaim itself only at such time as vascular tone was high, giving transient attacks of blueness, and necrosis being ultimately determined by a fresh thrombotic event. Pickering and I (10) have recorded a case (Case 17 of our report) in which a single attack of necrosis of the fingers in a warm-handed girl was followed by attacks of cyanosis; this case supports the idea just expressed.

The cases exhibiting necrosis or healed necrosis of the fingers here recorded strongly support the suggestion which Pickering and I put forward that thrombosis in these cases is a main determinant of necrosis of digits, not only in its massive form, but in the instance of the minute necrosis of the finger tips.

From the cases of Tables 2 and 3 it is clear that in patients who display necrosis of the finger tips, with or without healing, the digital arteries

TABLE 3.

Cases from past records.

Case.	Sex and age.	Attacks of discoloration	Frequence.	Provoked by	Nutritional changes.	Vascular lesions and other notes.
1. Gallavardin and Bernheim (5)	F. 68	4 y.	Frequent		Recurring small whitlows; then gangrene of half a finger.	Amputated finger, limited examination.* Digital arteries almost filled by fibrous tissue, lying within the main elastica and containing newly formed blood vessels. One vein showed appearances of endophlebitis.
2. Gagel and Watts (4)	M. 48	3 m.	Frequent		Bilateral gangrene, ends of fingers and toes (6 w.)	Carcinoma of pylorus. Endarteritis obliterans† with thrombus formation in small vessels of fingers.
3. Spurling, etc. (13)	F. 25	16 y.	—	Cold	Recurrent gangrene of ends of many fingers (2 y.)	Amputated fingers. Digital arteries showed intimal thickening (of grade here called moderate). Lumen of one "filled with an amorphous débris." Small artery plugged with clot. No digital artery described as obstructed, but examination limited. A sister affected. Patient's feet and face involved.
4. Lewis and Pickering‡ (10, Case 20)	F. 73	5 y.	Frequent	Cold air	Bilateral gangrene of ends fingers (4 m.)	Digital arteries of necrosed and un-necrosed (discoloured) fingers show moderate or conspicuous intimal hyperplasia. All lumens plugged by thrombi, recent, hyaline or recanalised (hæmosiderin). Toes also discoloured.

5. Dolio (3)	F. 31	Single attack of bilateral discoloration and necrosis of tips of fingers (7 m.)	Fingers amputated. Arteries (limited examination) showed fibrosis and thickening of intima. Very small lumens, some occluded by more or less organised thrombi. Also endophlebitis.
6. Nonne (12)	F 30	Single attack of discoloration of hands with ulceration of one finger.	Amputated finger. Small arteries show high grade of endarteritis† and periarteritis; very small lumen, or vessel occluded. Limited examination.
7. Cunningham (2)	M 33	5 yrs after an attack in toes, gangrene of two middle fingers (a few weeks).	Amputated finger. Digital arteries showed conspicuous intimal thickening, uninvaded by elastic and containing new blood vessels (presumably recanalised clot). Limited examination.
8. Lewis and Pickering‡ (10, Case 10)	F 67	Single attack of discoloration (bilateral) and necrosis of of tips of fingers (3 m.).	Digital arteries of necrosed and un-necrosed (discoloured) fingers showed moderate general intimal hyperplasia. All lumens plugged by thrombi, recent, or recanalised (haemosiderin). Toes also discoloured.

Bret and Chalier's (1, Case 1) (possibly thromboangeitis obliterans) and Grenet and Georges' (6, Case 1) are not included in this table, because the former includes no report of the digital arteries and the latter no clinical details.

* Limited in the sense that digital arteries were not examined in sufficient length to exclude actual obliteration of lumen at some point.

† The word "endarteritis" is used as a description by the authors. It is perhaps unnecessary to point out that this term is employed in a loose way in contemporary writing and often to signify no more than intimal thickening.

‡ Before including these two cases I have re-examined the sections of the vessels, so that the descriptions may be strictly comparable with those of cases in Tables 1 and 2.

may be assumed to be the seat of gross obstructive or occluding disease. In such the presence of this grade of disease is very possibly, if not probably, adequate to bring about arrest of bloodflow to the fingers whenever the affected arteries contract in normal response to changes of vasomotor tone or in direct response to cold. The structural disease in all such cases may be regarded as constituting what I have previously termed (9) the " local fault."

Origin of the arterial disease. There are two groups of cases presenting trainsient attacks of discoloration, the one without and the other with such arterial disease as might be held accountable for the attacks. Are these two types merely stages in the progress of one disease, or are they to be regarded as separate maladies ? Briefly, are we to regard the arterial disease as secondary to transient attacks of arrested circulation ? It is still impossible to return a completely satisfactory answer, though the position is becoming clearer. In the first place it is necessary separately to consider change in the vessel wall and the thrombotic accidents that are superimposed.

The main change found in the arterial wall in " Raynaud's disease," is hyperplasia of the intima ; but this change is no more displayed in these subjects than it is in many of those whose hands have always been warm and who give no history of attacks of discoloration. Thus the disease *of the arterial wall* is not to be ascribed to the attacks. In considering this relation from a theoretical standpoint it should be remarked that so long as the fingers are free from necrotic changes the arrest of bloodflow occurs only for relatively short periods, with much longer intervening periods in which the bloodflow is normal.

Occlusion of a vessel when arising from organic cause is generally the result of thrombus formation and this is an abrupt affair. It may be reasonable to suppose that in certain instances thrombi form in vessels the endothelium of which has been damaged by previous, prolonged or repeated, loss of blood supply. But in other cases this assumption is a difficult one to make ; there are cases in which the history of attacks of discoloration of the fingers has been associated from the beginning with necrosis ; cases where the first attack is one of prolonged discoloration ending in necrosis, and where this is followed by transient attacks of discoloration ; there is the fact that cases presenting fresh or healed necrotic lesions are cases in which previous attacks of discoloration are nearly always stated to have occurred over a relatively short period, a period of a few months or a few years, rather than a history of attacks since childhood. Such considerations lead to the belief that a proportion, perhaps a considerable proportion, of cases experiencing necrosis, are cases in which the whole train of symptoms from attacks of discoloration to ultimate necrosis, is determined by an initial thrombotic event.

PART 3. SCLERODERMA WITH RAYNAUD PHENOMENON.

These cases are discussed separately because it is possible that the primary mischief is in the tissues surrounding the vessels and that the vessels are secondarily affected. I have thought it important to record my own case, though many have been recorded previously, so that descriptions of the different conditions as given by the same observer may be available for comparison.

Case 7. Scleroderma (diffuse) with Raynaud's phenomenon. A.B., a married woman of 56 years was admitted to hospital in October, 1936, complaining of her fingers and toes. This was her history.

She first noticed attacks of discoloration of fingers and toes 9 years previously, the discoloration appearing on exposure to cold. The digits would become white and numb and later blue. During the same period stiffness of her fingers and of other joints of upper and lower limbs had developed. For a few years sore places had been forming on her fingers, and sometimes on ears and nose, in cold weather or following injuries. She had suffered from pain in the fingers and from worse pain in the toes. Latterly she had been breathless.

On examination she presented the characteristic picture of diffuse scleroderma. The skin of the whole face was smooth, shiny and stiff. The cheeks were covered with telangiectases; the naso-labial folds were lost, movements of the face were limited, the eyelids could only just be closed. The skin of all the limbs was similarly affected, the condition growing worse when traced distally. Movements of fingers, wrists, elbows, shoulder, and knee joints, were limited, apparently chiefly through the tenseness of the skin. The 4th and 5th left fingers were contracted into the palm, and the palm was thickened as in Dupuytren's contracture. Blood pressure was 170 systolic and 100 diastolic. The skin was much pigmented. The finger tips showed small scars.

A bilateral lumbar sympathectomy was done successfully. But during the second stage of a bilateral cervical sympathectomy the patient developed pneumothorax and did not recover.

Post-mortem. The cause of death was found in bilateral collapse of the lungs. The aorta and main arteries showed little atheroma, the main veins were normal.

Sections of the skin of a finger showed little change in the epidermal layer, the subpapillary layers of the skin were unusually rich in dense connective tissue and contained many small lymphocytic infiltrations. Sections of the ulnar and radial arteries showed medial sclerosis. The radial artery presented a moderate general hyperplasia of the intima, pervaded by coarse and fine elastic fibres, arranged in parallel laminæ. The ulnar artery showed more conspicuous but irregular intimal thickening, with much reduced lumen; the thickened intima presented the same appearance in its outer layers as those seen in the radial artery, but in the

ulnar the thickening was supplemented by internal and irregular pads of
sparsely nucleated connective tissue, with little elastic tissue. The main
palmar artery was in the same state. The digital arteries were all examined
in their whole length in the left 2nd and 4th fingers. From tip to base the
arteries of finger 4 showed conspicuous intimal thickening by tissue pervaded
by parallel wavy layers of elastic in its outer parts and by sparsely nucleated,
irregularly arranged, connective tissue (with or without fine elastic fibres)
within. One artery was replaced at the end of the finger by a number of
small ones, themselves showing intimal thickening (Fig. 14). Near the tip
of this finger the connective tissue around the vessels was very dense and the
medial coats of the digital artery was very fibrous. The vessels presented
the same appearance whether surrounded by fibrous tissue or not. Vascularity
of the adventitia was often conspicuous, with occasional but slight perivas-
cular infiltration. In the 2nd finger the changes were similar but more
advanced, both arteries had been closed (Fig. 13) chiefly by similar intimal
thickening, though here and there with tissue containing vacuolated cells
centrally ; these arteries were also surrounded by dense connective tissue,
sometimes penetrating and almost destroying the medial coat. The elastic
lamella was broken in places in most of the arteries examined. Hæmosiderin
was seen in the walls of none. The circulation was carried on in the 2nd
finger by a number of small muscular arteries, themselves showing intimal
thickening.

Summary. The case was one of diffuse scleroderma with Raynaud's
phenomenon developing simultaneously for 9 years, and associated in later
years with sore places on the fingers.

Examined histologically the ulnar artery, the palmar, and digital
arteries present conspicuous intimal thickening. The outer layers of thickened
intima contained parallel wavy elastic fibres, but the inner layers consisted
mainly of connective tissue.

The changes in Case 7 are similar to those previously recorded by Matsui
(11) in 5 Japanese women suffering from scleroderma and Raynaud's
phenomenon. As in our case, little change was found in the radial artery,
but gross changes in the digital arteries. Matsui described the media as
hypertrophied, and sometimes as sclerosed. The adventitia was less often
described as sclerosed, sometimes as infiltrated or surrounded by lympho-
cytes. In some vessels the intima was unthickened, but in the rule
conspicuous thickening with almost complete obliteration of the lumen
occurred, the thickening consisting of connective tissue and elastic fibres.
In two cases, out of four which presented necrosis of finger tip, organising
or organised thrombi were found in the digital arteries. The descriptions
of the digital arteries for the 6 cases are summarised with chief clinical details
in Table 4. These cases allow us to say that in scleroderma with Raynaud
phenomenon it is the rule to find conspicuous intimal thickening of the
digital arteries, in which increase of connective tissue plays an important

part. In the lumens, organising thrombi are not uncommon. Matsui made it clear from past records and his own observations that similar changes in the arteries occur in most of the tissues of the body and are not peculiar to the fingers or even to the skin of these patients.

TABLE 4.

Diffuse scleroderma and Raynaud's disease.

Case.	Sex and age.	Died of	Duration of symptoms in years.	Gangrene of finger tips.	Digital arteries.
Matsui's Case 1	F. 17	Heart failure	2	Dry	Organising clots (hæmosiderin) attached to walls.†
„ 2	F. 48	„	5	Small ulcers	Greatly thickened intima (collagen and elastic) ; lumen almost closed. Media thickened.
„ 3	F. 19	Pulm. tubercle	7	Dry	V. great intimal thickening (collagen and elastic).
„ 4*	F. 25	Pericarditis and tuberculous glands	1	Dry sores with scabs	Organised thrombi in places.†
„ 6	F. 33	—	3	Not mentioned	Media fibrosed. Conspicuous intimal thickening (collagen and elastic) ; lumen almost closed.
Own Case 7	F. 56	Operation for sympathectomy	9	Scars of healed ulcers	Sclerosis of adventitia and media. Conspicuous thickening of intima (connective and elastic tissue) obliterating lumen in places.

* In this case the scleroderma was not universal, but confined to the upper limbs, where it was advanced.

† Intima of digital arteries not mentioned, but it showed the usual thickening in other cutaneous arteries.

PART 4.   CAPILLARY PULSATION.

In 1924 I recorded observations (8) upon capillary pulsation of the finger tips produced by soaking the hand in hot water. In a series of 39 normal subjects of different ages, this capillary pulse was found to decline from about 40 years and often to fail after the 60th year was passed. It was suggested that the presence or absence of capillary pulsation in the fingers of the heated hand might serve to test the condition of the vessels of the fingers. The present observations allow this suggestion to be examined on an anatomical basis.

Capillary pulsation failed to be elicited in one subject (No. 18) of Table 1. The condition of the corresponding digital arteries in all eighteen subjects can be gathered from this table.   It would appear that slight or even moderate general proliferation of the intima is consistent with distinguishable capillary pulsation.   The most advanced instance of disease in those presenting the sign was found in No. 15.   In the 4th finger of this man during life capillary pulse was just clearly distinguishable when the hand was heated.   Both arteries showed moderate intimal hyperplasia at the base of the fingers and more conspicuous thickening (illustrated by Fig. 15) distally.   The grade of intimal thickening found in subject 18, in whom capillary pulsation could not be detected was very similar.   Of the Raynaud's cases in Table 2, three showed capillary pulsation during life.   It was shown by one patient (Case 3) in whom the intimal thickening was uniformly moderate.   The cases in which it failed to appear were those with nutritional changes in the fingers and for the most part plugged vessels (Cases 4, 5 and 6 of Table 2, and also Case 7 of Table 4).

We are in a position to conclude that failure to elicit capillary pulsation by soaking the hand in water at 46° up to 10 min. indicates a considerable reduction or obliteration of the lumen of the digital arteries.   The sign may be so used provided that there is a normal pulse pressure, pulses at the wrist, and that the pulsation is searched for carefully by one who is accustomed to elicit it.

CONCLUSIONS.

1.   In warm-handed adults, used as controls, the digital arteries usually show distinct intimal thickening, and this increases with age ;   general thickening is the rule from 50 years onwards.   The thickening consists of hyperplasia of the intimal tissues.   After 60 years conspicuous thickening may be found.

2.   In cases of intermittent spasm of the digital arteries, exemplified by the mildest form of so-called " Raynaud's disease," there is no more intimal thickening than is to be found in the arteries of warm-handed people of similar age.   In a case of the same type but of greater severity more intimal thickening than is usual was found, but it was no more than that found in many subjects who present no symptoms.   Attacks of discoloured fingers in these cases must therefore be ascribed to over-action of the muscular wall.   Hyperplasia of the media has not been found in any of them ;   the digital arteries are normally very muscular.

3.   In 2 cases of intermittent spasm presenting the scars of small healed necrosis of finger tips, obstructive disease of the digital arteries was discovered.   The arterial wall generally presented intimal hyperplasia ; the lumens were conspicuously reduced or actually occluded by new cellular tissue, or by recent or organised thrombus.   These conspicuous lesions were present in scarred and unscarred fingers.

4. In cases of intermittent spasm presenting unhealed necrosis of fingers thrombotic obstruction of the digital arteries is the rule ; it is seen in various stages of organisation.

5. In cases of bilateral discoloration and necrosis of fingers, without previous attacks of discoloration, the predominant lesion is again thrombus in various stages of organisation. Similar lesions are found in necrosed and un-necrosed fingers of these cases (*see* Lewis and Pickering).

6. It is suggested that in a proportion, perhaps a considerable proportion, of cases experiencing attacks of discoloration leading up to necrosis of the fingers, the whole train of symptoms is determined by an initial thrombotic event.

7. Evidence has not been found that attacks of digital arterial spasm lead up to thickening of the medial or intimal coats of the digital arteries ; very possibly it may lead up to thrombotic events.

8. In diffuse scleroderma, presenting attacks of discoloration of the fingers, there is conspicuous or occluding disease of the digital arteries. In the intimal thickening, hyperplasia of the original tissues and connective tissue growth play conspicuous parts ; organising thrombi may be found in the lumen.

9. In the presence of a full radial pulse, failure to elicit any capillary pulsation at the tips of the fingers, by adequately heating the hand, indicates a considerable reduction or obliteration of the lumen of digital arteries by structural disease.

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Fig. 1.
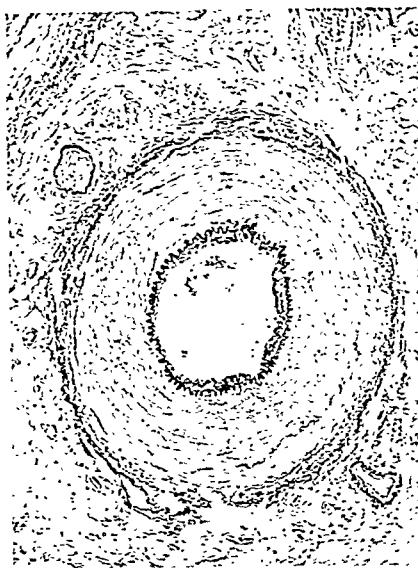

Fig. 2.

Fig. 1. (× 70). No. 2 of Table 1. A woman of 24 years. Warm hands. Digital artery near end of 4th left finger. Almost if not quite normal. Weigert's stain. Fig. 2. (× 70). No. 5 of Table 1. A man of 39 years. Warm hands. Digital artery near end of 4th left finger. Slight thickening cf intima. Weigert's stain.
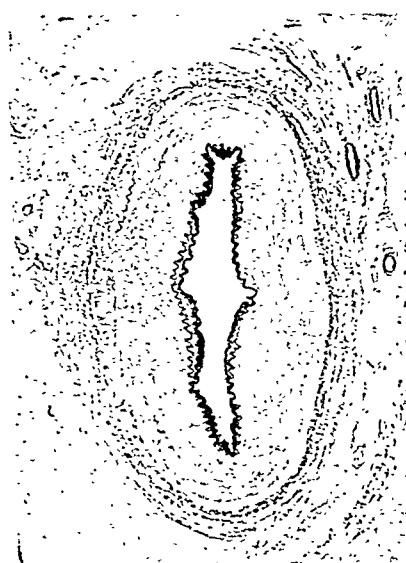

Fig. 3.
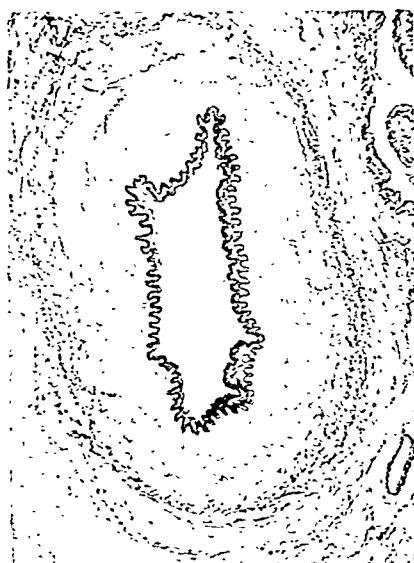
Fig. 4.

Figs. 3 and 4. (× 70). Case 2, aged 58. Raynaud's disease, mildest form. Digital arteries at base of right 2nd finger (Fig. 3) and at middle of right 4th finger (Fig. 4). There is slight patchy thickening of the intima. Weigert's stain.

Fig. 5.

Fig. 6.

Fig. 5. (× 70). No. 9 of Table 1. Warm hands. Digital artery near end of left index finger to illustrate "moderate general" hyperplasia of the intima. Weigert's stain.
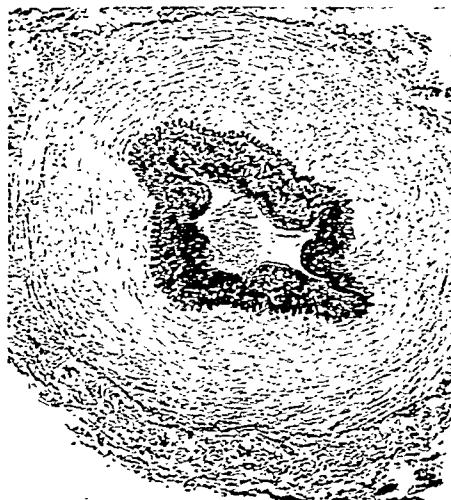
Fig. 6. (× 70). Case 3. Raynaud's disease, aged 44, uncomplicated. Digital artery from the right index finger. Adventitia and media show little abnormality. The intima is moderately and generally thickened. Weigert's stain.
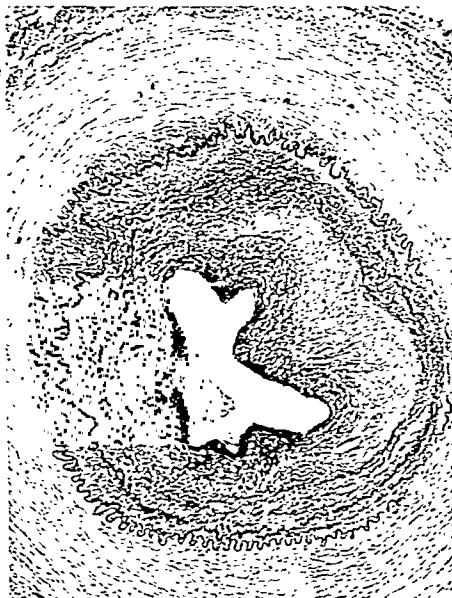
Fig. 7. (× 70). Case 3. Palmar artery showing conspicuous thickening of intima. The elastic lamella is broken to the right. Weigert's stain.

Fig. 8. (× 70). Case 5. Raynaud's disease, severe form. Digital artery from base of 4th right finger. The lumen obliterated by almost homogenous, sparsely nucleated, connective tissue. Hæmatoxylin and eosin.

RAYNAUD'S DISEASE AND PREGANGLIONIC SYMPATHECTOMY*

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EIGHT years ago in reporting the effect of cervical sympathetic ganglionectomy in two patients (one of them a case of Raynaud's malady), Landis and I recorded (5) the fact that within a few days of the operation, the vasodilatation consequent on loss of vasomotor tone declines from its full degree. Physiologists had already remarked (1) upon increase in the tone of minute vessels after sympathectomy in animals ; we added evidence that the small arteries are also affected. This was, so far as I am aware, the first evidence offered to show that the full effect of sympathectomy in man is not maintained, and this owing to the intervention of a local compensatory factor. It did not escape us at the time that this observation was relevant to the after-histories of patients with Raynaud's disease treated by sympathectomy. But I was concerned with the mechanism of the attack of spasm in digital arteries, believing I had found evidence that it was not primarily the result of abnormal vasomotor tone, as Raynaud had believed it to be. An argument used for this view was that patients would display attacks of loss of circulation to the fingers even after sympathectomy. I recognised that an attempt might be made to explain the recurrence of attacks after sympathectomy by attributing them to the interposition of the compensatory factor ; but refrained from discussing this point because the reply to such an argument seemed so clear. For if recurrence was not due to an original and abnormal local factor, as I supposed it to be, and if it were due to a new factor of compensation arising out of sympathectomy, these attacks of Raynaud's phenomenon should occur equally after sympathectomy, whether the cases operated upon were originally cases of Raynaud's disease or not. But this has not been shown. On the contrary, in so far as my own experience is concerned, only the case originally presenting the Raynaud phenomenon displays it after operation. To my mind it is conclusive that, if any case of Raynaud's disease presents attacks after ganglionectomy, the attacks are the result of a local fault both before and after operation.

Since our paper was written, the regain of vascular tone following sympathectomy has won increasing notice and especially through the interesting work of Telford (8), Smithwick (6) and White (7, 9, 10). These surgeons, basing themselves largely on new experimental work by Freeman, Smithwick and White (2), and by Grant (3) have become impressed by differences in the reaction of the vessels deprived of their sympathetic nerve supply by preganglionic interruption and by degeneration; they have devised operations of preganglionic sympathectomy in man (Telford and Smithwick); and they have concluded from these direct clinical observations that removal of the cervico-thoracic sympathetic ganglia is less effective in relieving Raynaud's phenomenon than is preganglionic section of the same sympathetic supply. I am prepared to accept clinical evidence for this conclusion. The preganglionic operation has the merit not only of producing, so it would seem, the greater vasodilatation, but in avoiding the disfigurement of Horner's syndrome (Telford (8)), the sympathetic motor fibres to the eye passing from cord to ganglion seemingly through the white ramus of the 1st thoracic root. The new surgical procedure is rapidly replacing ganglionectomy, the results of which are now very generally admitted to have been far from fully satisfactory. But the reports of preganglionic section, an operation which I welcome as more promising than ganglionectomy, are apt to convey too favourable a regard for this treatment. They are providing also—and this is a matter with which I am at present more especially concerned—a basis from which it may again be argued that attacks of Raynaud's phenomena are primarily the result of abnormal vasomotor nervous tone. But it is not true that preganglionic section of the vasomotor nerves to the upper limb always, or nearly always, prevents subsequent attacks of loss of circulation to the affected fingers, as the following observations quite clearly show. These observations concern six cases of Raynaud's phenomenon treated by preganglionic sympathectomy and examined before and shortly after operation. The cases are unselected in the sense that they have been the first six cases submitted to this operation that have been available for my tests.

The sympathectomies were all done by my colleague Mr. Gwynne Williams, to whose co-operation I am much indebted and who has kindly consented to their publication.

Case 1.—Severe Raynaud's disease with nutritional changes. Excision of left 2nd dorsal sympathetic ganglion; section of right cord below 2nd dorsal ganglion and reflection of cord upwards. Subsequent attacks in both hands.

E.W., a gardener of 33 years came under observation in October, 1937. As a lad he used to suffer from chilblains on the feet. His hands gave him no trouble before his 26th year, when he was laid up in bed with influenza. After getting up he noticed that his fingers often went white from tip to base. These attacks continued, increasing in frequency, the fingers becoming blue

or white in colour regularly on exposure out of doors or after immersing the hands for long in cold water. The discoloration would disappear in a warm room and the hands become very red and warm. From his 31st year there had been pain in certain finger tips, especially in the left and right index. Sore places with a little discharge appeared on several finger tips a year later, and these were very painful. Hæmaturia or discoloration of the face had not been experienced. Apart from coldness of the feet he made no complaint of them.

On examination in an outpatient department, all the digits (1 to 5) of both hands were blue (tint XIV or XV) to their bases, and the dorsal surfaces of the hands were patched with blue and red. The fingers seemed a little swollen, especially in the region of the proximal interphalangeal joints, and their flexion was imperfect ; the skin of their dorsal surfaces was, however, normally mobile. The tips of a number of fingers presented tiny depressed scars ; thus, on the left side these scars were present on the 3rd and 4th, and were numerous on the 2nd finger tip. The 5th finger tip was distinctly shortened and the nail narrowed and bent. Several of the nails were brown in colour and a little roughened and malformed. The skin beneath the majority of the nails was heaped up and adherent to the nail.

After immersion in warm water the hands became bright red all over. This redness remained when the hands were immersed in water at 44° but the tips of fingers 2 to 5 on both hands then became a little cyanosed (tint XI). When thoroughly heated no capillary pulsation could be found anywhere in the hands and no pulsation could be detected in the digital arteries. The radial pulses were full.

Immersion of a hand in water at 15° to 18° soon induced cyanosis, the whole hand becoming fully cyanotic (tint XV) to the wrist and large areas of the same discoloration appearing as high as the middle of the forearm within 20 min.. If a single finger of the warmed hand was immersed at 15°, this finger became fully cyanotic.

Sympathectomy was done on the left side, an inch of the cord, including the 2nd dorsal ganglion being excised on 14th December.

The patient was seen 19 hours after the operation. His pupils were equal ; the right but not the left side of the face and neck was sweating a little. The body was covered with sweat, but only as high as the 2nd intercostal space on the left side and in this space it was slight. The left arm and hand were quite dry, the axilla on its inner side only being moist. The right hand and arm were moist. His temperature was 100·2. The hands when withdrawn from under the bedclothes were equally and fully warm. Both hands were of a bright red tint, but the left hand and forearm were paler than the right, and the veins were more distended. Capillary pulsation could not be detected in either hand, and a digital pulse only at the base of the 5th right finger. The left hand was immersed in water at 15° and was maintained in the water, which rose to 18° during the next 45 minutes. Discoloration of fingers began to appear in 20 minutes, and

by the end of the immersion, the 1st, 2nd, 3rd and 5th fingers had become fully cyanotic to their roots. The 4th finger was less cyanosed. The whole of the back of the hand was almost fully cyanosed.

A right sympathectomy, division of the cord below the 2nd dorsal ganglion with reflection of the cord upwards into the scalenus, which necessitates section of the rami, was done on February the 8th. The right hand was hot and dry when examined next day. On the 12th (4th day) the hand was immersed in water at 17°, and the fingers became fully cyanotic (tint XV) to their roots within 11 minutes.

Although attacks could be provoked easily in both hands, they were usually warm and free from spontaneous attacks of discoloration. Their condition was much improved from their state before operation ; the man returned to hospital after the first operation, asking that the second should be done.

Comment. The operations that were done are to be regarded as complete sympathectomies of the limbs, with the exception of such skin of the axilla as may have been supplied by the 3rd ganglion, and preganglionic with the exception of the supply of the 2nd ganglion.

This first case shows that in Raynaud's malady attacks of loss of circulation to the fingers may be induced within a day of preganglionic sympathectomy. Attention is to be drawn to the fact that the affected fingers presented nutritional changes, and that evidence was obtained indicating structural disease of the digital arteries. Failure of sympathectomy to prevent attacks of Raynaud's phenomenon, clearly forbids us from ascribing the original attacks to overaction of vasomotor nerves. It is manifest that the attacks were due to an abnormality of the digital arteries. There was in fact independent evidence that these were diseased. The view can be taken that such digital arterial disease as this is entirely secondary to long-continued or repeated vascular spasm, and consequently is not to be regarded as an essential factor in the primary mechanism of a transient vascular closure. There is, however, insufficient evidence to support this conjecture ; but even were it sound it would not alter the fact that in cases of this kind as they come before us, the local fault must still be regarded as predominant. But, since evidence derived from cases presenting evidence of arterial disease is regarded by some as inapplicable to the malady in general, in that the arterial disease may be thought to arise as a secondary effect of a distinct primary fault, I shall describe other cases.

Case 2. Raynaud's disease. Division of left sympathetic cord below 2nd dorsal ganglion, and excision of right 2nd dorsal sympathetic ganglion. Subsequent attacks.

A.L., a shop assistant of 43 years came under observation in June, 1936. She gave a history of cold extremities and chilblains on the hands and feet in childhood, but of good health until the last few years. From the age of 39 years she experienced attacks in which the fingers became waxy white

in colour. The 4th and 5th fingers were the most often affected in each hand, but on each the 2nd and 3rd fingers were also affected, but not the thumbs. Blueness of the fingers often followed whiteness and, after recovery, the fingers became very red and a little swollen. Recovery was associated with considerable pain starting at the tips of the fingers. The attacks were provoked by exposure to cold ; they were confined to the winter time. A small blister appeared on the left 4th finger in March, 1936, following an injury by a small splinter ; it healed slowly. Numbness and discoloration of the toes had also been noticed ; her face was unaffected. There was no family history of similar disorders. She had experienced no hæmaturia.

Attacks of characteristic discoloration were seen. The skin of the fingers of both hands was normal in texture and mobility. The nail of the 4th left finger was discoloured, and near its base on one side a flake of discoloured skin was becoming detached. The radial pulses were full, the ulnar pulses palpable.

The left sympathetic cord was divided between the 2nd and 3rd dorsal ganglia on June the 17th, 1936. Next day the left hand was pink, warmer than the right, and quite dry ; but within 5 days abnormal blueness and coldness of this hand were noticed. The right sympathetic cord was exposed and an inch of it removed, including the 2nd dorsal ganglion on October the 8th, 1936. Next day the right hand was warmer than the left although exposed for many minutes outside the bedclothes. Within 4 days attacks of coldness of the right hand and blueness of the 4th and 5th fingers were noticed.

Examined on December the 11th, the woman stated that although her hands had been improved, she still experienced attacks, fingers 4 and 5 of the left hand particularly becoming numb and white for about 5 minutes on cold mornings. Her eyes were normal in appearance. The hands and axillæ were quite dry. On heating the patient, sweating occurred on the trunk to the level of the 3rd intercostal spaces on the two sides ; the arms, and head and neck did not sweat in any part. No scarring could be found on any finger ; the crumpled ridge on the 4th left finger nail was growing off. Flexion of all the fingers was quite free. When the hands were thoroughly warmed, capillary pulsation was seen in all fingers but the 4th and 5th right, and the 3rd left. When they were immersed in water at 18°C., the tips of all the digits of both hands, except the right 2nd and 3rd, acquired a deep cyanotic tint (tint XIV or XV) within 20 minutes. The hands continued in their improved condition until November, 1937, when the 3rd right finger became ulcerated and a large area of superficial necrosis occurred. Seen in February, 1938, this finger was still flaking, though nearly healed. Her hands were quite dry and she spoke of their having been so since the operation. The left hand was cool, the right cold ; and the last three fingers of the right hand deeply cyanosed.

Comment. The operations are to be regarded as complete sympathectomies of the upper limbs, with the exception of such skin as might be

supplied by the 3rd ganglion to the axillary region ; and preganglionic with the exception of the 2nd ganglion. These sympathectomies led to successive improvement of the two hands, but did not prevent attacks of discoloration occurring within a few days of operation.

In this case, unlike the last two, there was no clear evidence of nutritional change in the skin, a small lesion which had occurred on one finger being attributable to a well defined injury. It is proper to call attention to the fact that capillary pulsation was not provoked in two of the fingers chiefly affected, a common enough observation to make in cases of Raynaud's malady and one which must be interpreted as indicating significant structural change in the arteries of the corresponding fingers. Any remaining doubt that preganglionic sympathectomy may fail to cure, though it relieves, in cases of Raynaud's malady in which all the arteries to the fingers are without significant structural change is removed by the case now to be recorded.

Case 3. Raynaud's disease without nutritional changes. Bilateral excision of 2nd and 3rd dorsal sympathetic ganglia. Subsequent attacks.

L.W., a girl of 22 came under observation in June, 1936. She gave the history that, since the age of 14, her fingers and her toes became " black " and numb when she was cold ; that the attacks now occurred almost every day in winter time ; and that the discoloration started at the tips of the fingers and spread up the fingers to their bases. The right thumb and index finger were said to be most often attacked, but all were involved. Her hands were usually moist with sweat. She stated that she had never had any sores on her fingers at any time and on examination the fingers were without trace of scars. The skin of all the fingers was normal in texture and mobility ; the nails were all well shaped and quite smooth. Flexion of the joints of the fingers was normal. Capillary pulsation was observed in all finger-pads when the hands were warm. Thus, she was not only without nutritional changes in the fingers but free from detectable obstructive disease of the digital vessels. Typical attacks of discoloration could be induced by immersing the hands in cold water. There was no family history of similar affection. She had never seen blood in her water. Her face was unaffected.

The left sympathetic cord was divided on October the 13th above the 2nd and also below the 3rd dorsal ganglion, the corresponding piece of cord with these two ganglia being removed. Nine days later, on October the 22nd, a precisely similar operation was performed on the right side. The removal of two ganglia at the second operation was confirmed by their histological examination. Horner's syndrome developed on neither side. After recovery from the second operation, on November the 9th, it was found that on warming the body no sweating occurred on the body above the level of the third intercostal spaces anteriorly. Both arms were free from sweat with the exception of a small area on the lateral wall of each axilla, extending down the arm about 2 inches on the left, and 3 inches on the right side.

The operation on the left side was done on October the 13th. It is stated that on October the 14th, the left side of the face was warmer than the right, and was not sweating. When the patient was examined on October the 19th, the palpebral fissures and the pupils were equal in size ; the left face was drier than the right and the left malar process just perceptibly flushed. The left hand was found to be warmer than the right and paler in colour. Capillary pulsation was seen in all finger-pads and in all parts of the palm tested ; digital pulses could be felt in all fingers. This hand was dry, while the right was moist.

The operation on the right side was done on October the 22nd. The hands were examined day by day subsequently from the point of view of temperature, colour, and the strength of pulsation of the digital arteries ; the examinations being carried out on each occasion after the hands had been exposed equally for at least half an hour.

The dorsum of the right hand remained warmer than that of the left for eight days, as judged by palpation. By measurement with a thermo-electric junction the temperature difference was found to average about $\frac{1}{2}$°C. on the first day after the second operation, and about $\frac{1}{3}$°C. on the fifth. The fingers of the right hand were warmer than those of the left on the first day, and were subsequently of equal temperature or slightly warmer until the sixth day, after which time they were of equal temperature. The right hand remained deeper in colour than the left for the first three days; subsequently both hands were of equal colour. The digital pulses were usually found to be stronger on the right side for about the first eight days after operation. On the twelfth day they were equal and conspicuously weaker than immediately after operation. Capillary pulsation appeared equally in both hands whenever tested.

During the period of the patient's recovery from operation she was in hospital under daily observation and the weather was cold. The hands were usually outside the bedclothes. Before operation typical attacks of blue discoloration of the fingers occurred frequently in both hands. These attacks almost ceased after the bilateral operation, though on one occasion, November the 1st (10th day), deep cyanosis of the right index finger appeared; it was confined to the finger tip.

It was found to be still possible to cause abnormal cyanosis in most digits of both hands. Thus, on November the 3rd, 21 days after the left sympathectomy, the left hand was immersed to the wrist at 17°C.. After 17 minutes all digits of the left hand were found to be cyanotic (tints XII to XIV), the cyanosis extending from the tip of each finger over one or two phalanges. On the same day, 12 days after the right sympathectomy, the right hand was similarly immersed in water at 18°C.. After 19 minutes the 2nd, 3rd and 4th digits were cyanotic (tints XII to XIV) over the terminal phalanges, and 9 minutes later the cyanosis had spread to the tip of the thumb.

In February, 1938, this patient reported that there were no further attacks until December, 1937, when they returned in all fingers. An attack

of discoloration was present when she came, all fingers being deeply cyanosed at their tips. The hands were quite dry and she said that they had remained so since operation, though she had noticed a little sweating under her eyes when nervous or hot. After immersion in hot water the fingers all displayed very distinct capillary pulsation.

Comment. The operations are to be regarded as complete sympathectomies of the limbs, and preganglionic with the exception of such skin of the axilla and inner side of the upper arm as may be regarded as supplied by the 2nd and 3rd nerve. On neither right nor left side did the operation prevent the early recurrence of attacks, spontaneous and deliberately provoked, though the condition of the hands was improved. Observations seemed to show that the vasodilatation following operation declined during the first few days to reach stability about the 6th to 8th day after sympathectomy. This case was one of Raynaud's malady presenting no nutritional changes and no evidence of significant structural disease in the digital arteries.

The two cases which follow were of milder Raynaud's disease, in which, though no attacks could be induced after sympathectomy, the vessels could nevertheless be shown to react abnormally.

Case 4.   Raynaud's disease without nutritional changes.   Excision of 2nd and 3rd left dorsal sympathetic ganglia ; division of right cord below 3rd dorsal ganglion and of rami to 2nd and 3rd ; subsequent abnormality shown.

M.A., an unmarried woman of 34 years came under observation on December the 30th, 1937. She said that her hands and feet had been cold since childhood, with attacks of discoloration of her fingers. Chilblains occurred on her fingers during the last two winters. The attacks occurred many times a day in cold weather, all the fingers becoming bloodless at their tips, the waxy colour spreading to the proximal interphalangeal joint, and the fingers becoming numb. When hanging down, the fingers and backs of the hand became deep blue in the attack. When the fingers went white, they always became blue subsequently. Attacks were provoked by cold weather or by immersion in cold water ; they were more frequent when she was worried or upset. Warmth gave recovery with reddening and tingling. Her hands had never sweated much. She had no knowledge of attacks in feet or face ; her urine was always normal in colour. There was no family history of the affection.

The skin of the fingers was normal in every respect, mobile and unscarred; the nails were normal. When spontaneously warm, or after immersion in water at 43°, the skin was red, pulsation of digital arteries could be felt in all the fingers, and capillary pulsation was observed in all the finger tips. When cold, the fingers became fully cyanotic to their roots and cyanosis spread over the backs of the hands. The colour of the skin of the hands was unusually deep as high as the wrists. Exposure of a warm hand to the air of the ward (17°) or immersion in water at 16° to 18°, induced obvious cyanosis within 10 min., and the cyanosis was full within 20 min..

Left sympathectomy was done on January the 4th, the 2nd and 3rd dorsal ganglia being removed (confirmed histologically). Horner's syndrome did not develop, but when examined next morning, the left hand was hot and dry, the right blue and cold as usual after exposure. Tested at a later date, this patient was found to sweat in response to general warming, over the right face, neck and arm ; but sweating was absent from the corresponding parts on the left side. On the trunk sweating ended at the level of the 3rd dorsal spine behind and at the 3rd left rib in front, except for a little moisture here and there in the 2nd interspace. The left axilla was a little moist, and a narrow band of moist skin extended down the inner side of the upper arm to, but not beyond, the elbow.

On the 1st day after operation the vasodilatation in the left hand was maximal, the hand was red and hot ; its veins were dilated ; pulsation could be felt in all the fingers to the very tips, capillary pulsation was brilliant in all the finger tips. Immersed in water at 15° to 18° for 10 or more min. the redness was unchanged, and on taking out the hand it was found still to be hot. It was impossible to cool it much by such immersion. So the bloodflow to the arm was arrested, after depleting the skin of blood by holding it up,* and the arm cooled for 15 min.. On releasing the circulation reactive hyperæmia appeared throughout the hand, but its appearance was delayed on the backs of the fingers ; they were not clear for 30 sec..

On the 3rd day the hand was in much the same state, but pulsation could no longer be felt in the pads of the fingers, the hand had paled a little, and the size of the veins was a little less. By the 6th day the paling in the hand had increased. Tested on both these days, immersion at 18° failed to induce an attack ; but the same delay in the appearance of reactive hyperæmia was noticed in the cooled hand. On the 14th day the hand was again paler and the pulsation in its fingers lessened, though the hand was continuously warm and all finger tips still presented capillary pulsation. On this day, the hand being raised, the vessels were occluded and the hand then immersed at 16° for 10 min.. Release was followed by reactive hyperæmia ; but all the fingers failed to clear for 60 sec. ; the index finger remained fully cyanotic over its distal half for 5 min. and was not quite clear in 10 min.. Thus definite abnormality was demonstrated (repeated with same result on the 54th day), and especially abnormality of the 2nd finger, which the patient herself had regarded as her worst.

Right sympathectomy was done on January the 25th ; the cord was cut below the 3rd dorsal ganglion, and freed from all attachments as high as the inferior cervical ganglion ; its free end was stitched into the scalenus muscle. Next day the hand was much redder, was warmer, the pads of the fingers pulsated more and capillary pulsation was more distinct on the right than on the left side. The two hands were compared day by day and had come almost to equality by February the 7th, the 12th day after right

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* This should always be done in these tests.

sympathectomy. On the 12th day the right hand was immersed in water at 17° for 25 min., the circulation being arrested for the last 10 min. and then released. Reactive hyperæmia invaded the fingers slowly, all the fingers being completely cyanosed at the 5th sec., the index finger being completely cyanosed in its length at the 10th sec., remaining without circulation at its tip for 1½ min. (repeated with the same result on 33rd day). This abnormality was in harmony with the patient's statement that the index fingers had always been the first to become discoloured and the last to recover.

Questioned on June the 7th, her hands had shown no attacks. Her eyes were normal. On being heated, the trunk became wet to the level of the 3rd ribs ; hands, arms and neck remained dry. But sweating was profuse on the central parts of her face. The patient stated that after her operation, *but not before,* her face always sweated profusely when she ate sour things, (compare Wilson, *Clinical Science,* 1936, 2, 273).

Comment. The operations are to be regarded as complete sympathectomies of the limbs, and preganglionic with the exception of such skin of the axilla and upper arm as may be regarded as supplied by the 2nd and 3rd ganglia. Although no spontaneous attacks occurred afterwards, abnormal responses of the fingers of both hands were demonstrated, by immersing them in cold water, arresting and releasing the circulation, within 12 or 14 days of operation.

Observations seemed to show that the vasodilatation following operation declined to reach stability about the 6th to 12th day after sympathectomy.

Case 5. Raynaud's disease without nutritional changes. Excision of the 2nd and 3rd left dorsal sympathetic ganglia ; right sympathetic cord divided below 3rd dorsal ganglion. Subsequent slight abnormality shown.

E.S., a married woman of 36 years, came under observation in March, 1937. She stated that she had experienced attacks of numbness of the hands and feet since childhood, the toes becoming white, the fingers blue and later white. All her digits were affected. The discoloration would start at the tips of the fingers and spread to their bases. The attacks occurred on exposure in cold weather, but they were not invariably affected under seemingly similarly conditions. Worry was said to induce attacks. Recovery in response to warmth made the hands red, and later they would become swollen. The feet had been very painful, but she had experienced no local sores or painful flaking of the skin of her digits. The hands sometimes became discoloured quickly when she used cold water for washing. Her face had never been affected. She had not suffered from hæmaturia. Her mother suffered from similar but occasional attacks in her fingers.

On examination the texture of the skin of her hands was quite normal, the nails were well formed, the skin of the fingers normally mobile and presenting no trace of scar. The radial and ulnar pulses were normal.

Seen again in February, 1938, the condition of her hands was unchanged. After the left hand had been soaked in water at 44° for 2 min., very distinct capillary pulsation could be seen in all the finger tips and palm. Exposure of the hands on the bedclothes in a ward at 16° for an hour failed to induce an attack. Immersion of the left hand in water at 17° gave unusual duskiness of the fingers and backs of the hand (tint XIII) in 15 min.. The bloodflow to the hand was arrested and the immersion continued for 5 min., when the bloodflow was released. Reactive hyperæmia spread very slowly beginning in the hand in 20 sec., spreading to the 1st and 5th fingers at 1 min., but leaving fingers 2, 3 and 4 persistently and fully cyanosed.

Left sympathectomy, excision of the 2nd and 3rd dorsal ganglia (confirmed histologically) was done on February the 8th. The left hand was hot and dry next morning, pulsation being felt easily at the finger tips, and capillary pulsation being vivid in all of them. On February the 12th (4th day) the left hand was immersed in water at 17°, and the back of the hand and fingers were more than normally cyanosed (tints XII and XI) in 12 min.. After holding the arm up and arresting the bloodflow it was re-immersed in the cold water for 10 min. and the bloodflow released. The reactive hyperæmia occurred slowly, all the fingers being fully cyanosed to their bases at 10 sec. and the index finger failing to clear for 45 sec.. On February the 18th (10th day) in the same test, the time was extended to 85 sec., reduced on the 14th day to 30 sec., and extended on the 18th day to 110 sec.. In these last tests it was particularly noted that fingers 3 or 4 were always the last to clear ; these two fingers according to the woman had usually been the last to recover in spontaneous attacks.

Right sympathectomy was done on March the 8th, the cord being divided below the 3rd dorsal ganglion, stripped of all connection as high as the inferior cervical ganglion, and sown into the scalenus muscle. Next day this hand was hotter and redder than the left, and dry. The signs of vasodilatation became similar to those in the left in about 3 days. The reactive hyperæmia test of the cooled hand, as described for the other hand was done on the 6th day, the finger tips were still fully cyanosed 30 sec. after release and not completely clear till 70 sec. ; on the 8th day recovery took 90 sec. and on the 13th day 30 sec..

Sweating was induced on March the 30th. It occurred symmetrically on the trunk as high as the 4th rib or 3rd space in front, and as high as the 4th dorsal vertebra behind. The axilla became a little moist and a little dampness was detected on the inner aspect of the upper half of each upper arm. The rest of the skin of arms, head and neck was quite dry.

Comment. The operations are to be regarded as complete sympathectomies of the limbs, and preganglionic except for the supply from the 2nd and 3rd ganglia. Although no spontaneous attacks occurred afterwards, abnormal responses of the fingers were demonstrated within 4 or 6 days of operation.

The remaining two cases, the first of very mild Raynaud's disease and the last of retinitis pigmentosa are added as controls. In neither of these two cases could attacks of Raynaud's phenomena be induced before operation ; in neither could they be induced, and in neither was it possible to demonstrate abnormality in the reaction of the digital vessels to cold, after operation.

Case 6. Mild Raynaud's disease. Bilateral preganglionic sympathectomy. Inability to induce attacks before, or to show abnormality after operation.

G.M., an unmarried woman of 49 years was first seen on January the 1st, 1938. She gave a history of cold hands and feet since childhood and of chilblains on hands and feet for a few years. For 10 years she had experienced attacks of discoloration and numbness of the fingers. The attacks occurred occasionally after going out of doors in cold weather or after bathing in summer time. She had never noticed any effect from excitement or nervousness. Her skin, including that of her hands, was declared to be unusually dry. All her fingers would be affected, though the thumbs unusually. The discoloration, first blanching and later blueness, would start at the tips and spread sometimes as far as their bases. Warmth and rubbing would bring recovery with a feeling of pins and needles. There had never been any sore places on her fingers. Her feet were affected in the same way, but less severely. Her face was unaffected ; no history of hæmaturia. Her father's fingers became white and numb in cold weather.

On examination the skin of the fingers and the nails were quite normal in appearance, the fingers could be flexed fully. Her hands were always cold and dry when exposed to the air of the ward (temp. 18°). The pulses at the wrists were normal. Though watched for 10 days in cold weather, no attacks of discoloured fingers were seen, and attempts to induce them failed. Immersing a hand at 18° for 30 min., including 10 min. of arrested bloodflow, failed to close the vessels ; releasing the bloodflow, the fingers coloured to their tips within a few seconds.

On January the 11th the right sympathetic cord was divided by excising the 2nd dorsal ganglion. The corresponding hand afterwards showed vasodilatation, being warm and quite dry, and pulsation being felt easily in its vessels and capillary pulsation being vivid in the finger tips. This hand became paler, cooler, and its pulsation less during the next week. Tested on the 9th and 27th day, no attack of discoloration could be induced in it.

On February the 1st the left sympathectic trunk was divided by excising the 3rd dorsal ganglion, the cord above being cleared as high as the inferior cervical ganglion and stitched into the scalenus muscle. Next day this hand was redder and hotter than the right, and palpable and visible pulsation in it were greater. Horner's syndrome was not present. The two hands were compared daily and seemed to be equal in condition for the first time on the 6th day following the left sympathectomy.

After cooling the hand (right on 27th, left on 6th day) at 18° for 25 min., and arresting the circulation to the arms for the last 10 min., the flush of reactive hyperæmia invaded the fingers to their tips in 7 to 10 sec..

Case 7. Sympathectic ganglionectomy for night blindness in a warm-handed man. Subsequent inability to induce attacks.

S.R., a man of 26 years, always warm-handed, suffered from retinitis pigmentosa. For this condition the right middle and inferior cervical ganglia with the 2nd and 3rd dorsal ganglia were excised in January, 1937.

Examined in February, 1938, he was seen to have Horner's syndrome on the right side. His hands were equally warm, but the right was dry, while the left was moist. He stated that the right hand was colder than the left in cold weather. His hands were soaked in water at 17° for 15 min., when the right hand was seen to be distinctly paler and pinker than the left. Both hands were held aloft, the bloodflow to the arms was stopped, and the hands were re-immersed. After 10 min. of arrested flow the circulation was released. The fingers of the right (sympathectomised side) were quite clear in 12 sec., those of the left side were clear in 30 sec.. The right hand warmed up quicker than the left.

In using this case as a control to the reactive hyperæmia test used in other patients, it is to be noticed that the time taken for hyperæmia to pervade the sympathectomised cooled hand, 12 sec., is much less than that taken for it to pervade the normal cooled hand ; and that the time in the former is conspicuously less than usual in patients 4 or 5 in which this same test is relied on to display abnormality in the sympathectomised hand. The fact that this control was one of long standing ganglionectomy and not preganglionic section, does not lessen the strength of the argument.

Discussion.

The main observations of this paper are summed up in the accompanying table, in which six cases of so-called " Raynaud's disease " are arranged in order of decreasing severity. In Case 1, nutritional changes were obvious in the fingers. In Case 2, a small lesion had occurred on a finger that had been injured. In Cases 3, 4, 5 and 6 there were no nutritional changes ; in Cases 3 and 4 attacks were frequent in cold weather and could be induced easily by immersing the hands in cold water. In Case 5 attacks were very difficult to induce, and only by cooling with the bloodflow to the hand arrested. In Case 6 attacks could not be induced either by simple immersion or by immersion and arrested bloodflow.

Of the hands of these same cases after sympathectomy it is first to be said that in the period directly following operation, the effects of the treatment were striking. In all cases hands that were cold before operation became warm or hot afterwards. When, as was usual, attacks of discoloration had been present before operation, these attacks were abolished or much reduced in frequency and severity. The full vasodilatation seen on the day after operation is not maintained in these preganglionic operations, the declined occurring

Case.	Nutritional changes.	Capillary pulse, finger tips.	Attacks.	Sympathectomy.	Subsequent attacks.	Abnormality to cold and occlusion.
1.	+	None	Very frequent and easily induced.	L. 2nd D excised. R. Cord cut below 2nd D.*	Attacks easily induced 1st and 4th day.	
2.	Small lesion from injury.	A few fingers only.	Frequent in winter.	L. Cord cut below 2nd D. R. 2nd D excised.	Spontaneous, 5th day. Spontaneous, 4th day.	
3.	0	Present in all.	Frequent in cold weather and easily induced.	L. 2nd and 3rd D excised. R. 2nd and 3rd D excised.	Spontaneous in R. on 10th day. Induced in L. on 21st day.	
4.	0	Vivid in all.	Frequent in cold weather, worse when worried; easily induced.	L. 2nd and 3rd D excised. R. Cord cut below 3rd D.*	None could be induced (L. or R.).	Pronounced, 6th day. Distinct, 12th day.
5.	0	Vivid in all.	Frequent in cold weather, worse when worried; very difficult to induce.	L. 2nd and 3rd D excised. R. Cord cut below 3rd D.*	None could be induced (L. or R.).	Distinct, 4th day. Distinct, 6th day.
6.	0	Vivid in all.	Occasional in cold weather, and bathing. Not to be induced.	L. Cord cut below 3rd D.* R. 2nd D excised.	None could be induced (L. or R.).	No abnormality shown.
7.	Retinitis pigmentosa.		None.	R. ganglionectomy I.C.G. and 2nd and 3rd D.	None could be induced.	No abnormality shown.

* In all these the cord was stripped up to the inferior cerv. ganglion, dividing all branches, and sewn in the scalenus.

gradually over a period of about a week (*see* Cases 3, 4, 5 and 6, and compare Telford's statement (8)). Despite this change, the benefit remains obvious. Thus, the surgical treatment of the series was unquestionably a success ; this result is attributable however to the removal of normal, and not abnormal, vasomotor nervous tone. That this is the correct interpretation is clearly established for the series by comparing the reactions before and after treatment.

Despite conspicuous improvement, spontaneous attacks occurred, or attacks were induced without difficulty, in the first three cases. In the remaining cases attacks of persistent spasm were not induced, even though cooling was accompanied by temporary arrest of the bloodflow. But in Cases 4 and 5 evidence was obtained that. under the more thorough cooling of arrested bloodflow, the vessels entered a state of spasm, resistant for a longer or shorter time to reactive hyperæmia. In only one case (Case 6) could no abnormality be displayed after operation, but in this case no attacks could be induced before operation.*

Briefly the cases form a series in the degree of their abnormality before sympathectomy ; they form a series of similar order in the degree of abnormality after sympathectomy. Sympathectomy has lowered the whole scale of abnormality without appreciably changing the relative positions of the members of the series. Now this is just the result to be expected if the effect of sympathectomy is the withdrawal of normal sympathetic tone. It is not the result to be expected if the Raynaud's phenomenon is due to unnaturally exalted sympathetic tone ; for whether the exaltation of tone were great or small originally, it should be removed entirely by sympathectomy, and all the cases should subsequently be reduced to the same state. This does not happen, and we are forced to conclude that the morbid factor in these cases is not abnormal vasomotor nervous tone, but a local abnormality. In the most severe case or cases of the series this local fault may be largely, if not wholly, obliterative vascular disease. In the least severe cases the fault cannot. be so regarded, for as recorded in the article (4) which precedes the present one, cases of such mild type exhibit little or no more vascular disease than do warm-handed subjects of the same ages. It was in my first paper (1929) that the argument was first used that, since Raynaud's malady is not necessarily cured by breaking the sympathetic paths, it cannot be due to abnormal sympathetic tone. To this it has sometimes been replied that the cases of Raynaud's malady then cited from this point of view were severe cases. They were no more severe than cases described by Raynaud himself and regarded by him as the immediate result of abnormal sympathetic tone ; that the more severe cases are proving to have digital arterial disease is consistent with my views but not with his. But the present series of patients definitely extends the evidence of the non-

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* As this paper goes to press I have seen a case in which no abnormality was displayed after operation, but in which attacks could be induced with difficulty before operation ; such minor variations are to be expected and do not affect the general validity of the arguments used in this discussion. It is also to be added that Case 6 experienced an attack of blueness and numbness of the tip of the right index finger when cold on May the 16th.

curative effect of sympathectomy (and this preganglionic) to cases in which structural change in the digital arteries cannot reasonably be regarded as determining attacks.

Thus we are brought to reiterate the conclusion, so far as the pathogeny of so-called " Raynaud's disease " has been explored, that the vasomotor nerves are not at fault, but that the fault is a local one. While in some cases this fault may be in the form of structural change, in others it appears to be different (Cases 3, 4 and 5 are of particular significance from this point of view) ; possibly in these the apparently increased susceptibility of the vessels to cold is due to sensitisation of the vessels by some circulatory hormone ; possibly it results from some other local influence.

SUMMARY AND CONCLUSIONS.

1.　Six unselected cases of " Raynaud's disease " have been examined shortly after preganglionic sympathectomy. In three of these, attacks of discoloration of the fingers occurred spontaneously or were induced within a few days of operation. In one of these three cases the fingers presented nutritional changes ; in another of the three there was no trace of such changes. Of the remaining three cases, in only two could attacks be induced before operation (and in one of these with difficulty) ; in these two abnormality in the reaction of the fingers to cold could be shown after operation.

2.　Preganglionic sympathectomy does not bring the fingers of these cases to a common state ; it relieves in all cases, but a local abnormality remains, and this can be displayed in a measure that is related to the abnormality displayed before operation.

3.　The attacks in these cases of " Raynaud's disease " are not due primarily, as Raynaud thought, to excessive action of the vasomotor nervous apparatus. They are due primarily to a local fault ; this may consist of occlusive structural disease, or it may not. In the latter case the digital vessels appear to present increased susceptibility to cold, the reason for which still remains obscure.

4.　The full vasodilatation resulting from preganglionic sympathectomy, declines during a period of about a week following operation.

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THE MECHANISM OF LOCAL SWEATING IN RESPONSE TO FARADISM.*

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DURING experiments in this laboratory in which a faradic stimulus was used to produce hyperalgesia of the skin, I frequently observed that beads of sweat appeared widely surrounding the electrodes. The extent of the sweating area seemed to preclude the possibility that the effect was a direct one of the current on the sweat glands. Nevertheless, the fact that the sweating was always in direct relation to the electrodes suggested a local mechanism rather than a generalised sweat response which is known to follow a painful stimulus. This local faradic sweating seemed suitable for investigation along lines similar to those used by Lewis and Marvin for goose skin (2).

Method.

The extent of the sweating area has been indicated by the method of Minor (3). A mixture consisting of alcohol 90 c.c., castor oil 10 c.c. and iodine 1·5 g. is painted widely on the skin. When it has dried, finely powdered starch is blown over the painted area with an atomiser. Where sweating occurs the starch and iodine are brought into aqueous solution and the familiar dark blue coloration results. The area is photographed through a yellow filter for permanent record. (Fig. 1).

The extent of the sweating area produced by a faradic stimulus varies with the current strength employed. A very painful stimulus (Du Bois coil secondary 6·2 cm., 4 volt battery) continued for 2 minutes will produce an area of perceptible sweating about 8 cm. × 3 cm.. Sweating first appears about ½ a minute after the electrodes are laid on the skin. It occurs most intensely in their proximity, while the outlying parts show as scattered sweat pores. Faradic sweating can be obtained on all parts of the body, but the

outside of the forearm shows the reaction most distinctly. This part has accordingly been used in the majority of these experiments.

Nervous mechanism involved.

The participation of a nervous mechanism in faradic sweating may be proved by applying a strong faradic stimulus to a small anæsthetic area produced by the intra-cutaneous injection of 0·1 c.c. 2% novocaine. No sweating occurs in the surrounding normal skin, showing that the nervous pathways have been blocked by the novocaine. Normal sweating occurs when a stimulus is applied over a similar injection of saline and also on the re-application of the stimulus to the novocaine area after its sensation has recovered.

Pathway involved. The nervous pathway between the stimulating electrodes and the sweat glands might be conceived as being purely local in the skin, or involving nerves to and from the spinal cord. In the latter case no faradic sweating should be obtained when the central pathways mediating sweating are blocked.

Two adjacent nerves supplying areas of contiguous forearm skin are anæsthetised by the injection of 1 c.c. 2% novocaine in each one after localisation by a weak faradic stimulus. This arm is now painted with indicator, while the opposite arm is immersed in hot water. The whole body surface sweats except the anæsthetic area produced by nerve anæsthetisation (Fig. 3), showing that the central sweat pathways have been blocked in this region. The application of a faradic stimulus to the middle of this sweat free area now produces the usual sweat response, showing clearly that this is a local mechanism.

In accordance with this idea it is found that faradic sweating may be obtained on areas rendered anæsthetic by actual nerve section, always providing that the nerves have not had time to degenerate. The experiment may be conveniently performed on areas of cutaneous anæsthesia produced following the operation of nephrectomy, in which section of a cutaneous nerve in the skin incision is frequent. Within 12 hours of the operation a faradic stimulus put down in the middle of such an area produces a normal sweat response. This area and a control in the opposite loin are retested from day to day with the same stimulus. By the 3rd day sweating on the anæsthetic area has become less extensive and intense. A few scattered glands only respond by the 6th day and no response is afterwards obtained, while control skin gives full sweating. Degeneration of the local sweat nerves has thus become complete by the 6th or 7th day.

Type of local nerve mechanism. The uniform and extensive area of sweating produced by a faradic stimulus might suggest that it spreads through some kind of closely anastomosing nerve network. Yet this is not so because a novocaine barrier (0·2 c.c. 2% novocaine intercutaneously)

placed in proximity to the electrodes abolishes a segment from the sweating area (Fig. 2). This result is very similar to that obtained by Lewis and Marvin for goose skin (2). It suggests that the nerves are arranged in a plexiform manner rather than a network. Several plexiform systems of nerves are known to exist in the skin ; but the fact that the local sweat reaction disappears after sympathectomy shows that this reaction is mediated through nerves of the sympathetic system.

Sweating after sympathectomy. The experiment has been performed on a case of unilateral sympathectomy (ganglionectomy) for retinitis pigmentosa. Two years had elapsed since operation. A faradic stimulus of sufficient intensity to produce widespread local sweating is laid down on the normal arm. A precisely similar stimulus applied to the sympathectomised arm produces no sweating.

Effect of atropine on the local reaction. It has long been known that sweating in man, although mediated through sympathetic nerves, can be abolished by atropine which is usually held to paralyse parasympathetic nerves only. This seeming discrepancy has been recently cleared up by the work of Dale and Feldberg (1), who have shown that the sweat nerves are in the cat cholinergic in function, liberating acetyl choline at their peripheral endings. The nerves responsible for local faradic sweating in man likewise appear to be cholinergic in function since no sweating is obtained from an adequate stimulus in an atropinised subject (Fig. 4).

This is in contrast to its effect on the goose skin produced by faradism, which remains unaffected. It was shown by Lewis and Marvin (2) that the goose skin response to faradism was produced through a sympathetic plexus of nerves. It is clear however, that a separate system of nerves is involved in the two cases for they differ not only in their response to atropine, but also in their spacial distribution. Although in general they are both distributed around the electrodes, large isolated patches of goose skin are often seen without any attendant sweating, and *vice versa.*

Hyperalgesia and the vascular flare following a faradic stimulus likewise tend to be fairly uniformly distributed around the electrodes, but their outline does not coincide exactly with that of sweating.

Comment.

From the clinical aspect the main interest of these experiments is that they demonstrate the existence of a local sweat mechanism in the skin. I have frequently observed sweating surrounding ulcers of the skin, and sweating of a single finger also occurs with a terminal whitlow. Such instances of local sweating are probably produced through the local mechanism described.

CONCLUSION.

Sweating can be produced by direct stimulation of the sweat nerves in the skin ; the impulses pass through axons of a local sympathetic plexus. These axons appear to be cholinergic in function.

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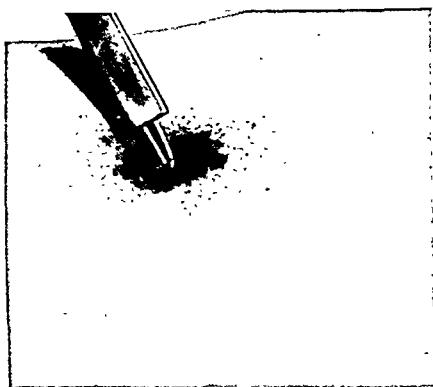
Fig. 1.
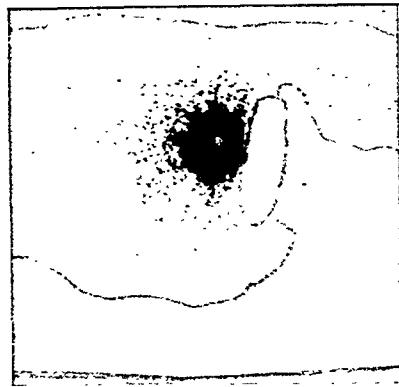
Fig. 2.

Fig. 1.   Local faradic sweating with stimulating electrode in place.

Fig. 2.   Effect of novocaine barrier.   The full area of sweating, and the barrier, are outlined.

Fig. 3.   Sweat free area outlined following anæsthetisation of forearm nerves *N*1 and *N*2 Local faradic response shown in middle of this area.
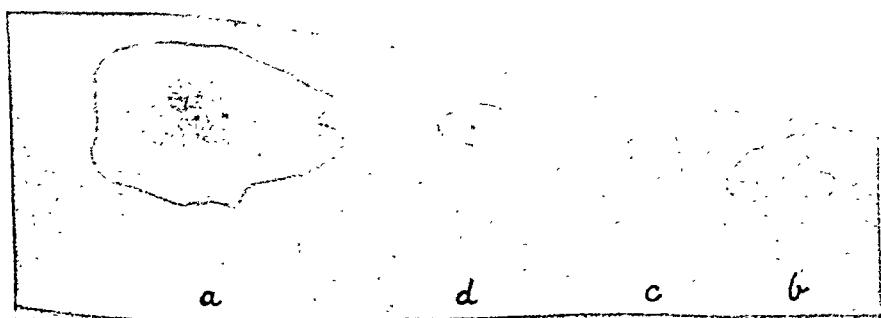

Fig. 4.   Effect of subcutaneous atropine g. 1/60.   (*a*) before atropine, (*b*) 20 min., (*c*) 35 min., (*d*) 90 min., after atropine respectively.

ACUTE ARTERIAL LESIONS IN RABBITS WITH EXPERIMENTAL RENAL HYPERTENSION.*

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THE importance of acute arterial lesions in the post-mortem differentiation of the malignant and benign forms of essential hypertension is now well recognised but the pathogenesis of these changes remains obscure. This paper describes the occurrence of acute arterial lesions in rabbits with experimental renal hypertension,† an observation which enables us to study experimentally some of the factors involved in their causation.

Methods.

Hypertension was produced in adult female brown rabbits by removing or destroying one kidney and constricting the other renal artery with a silver clamp ; or alternatively by constricting both renal arteries. Details of this method which is essentially that of Goldblatt and his colleagues (2) are to be presented in a paper by one of us and Dr. M. Prinzmetal and this histological study was made on tissues from the experimental animals there described. Arterial pressures in these animals were measured on the central artery of the ear. The various tissues examined were fixed in saline formaldehyde and the sections stained with Ehrlich's hæmatoxylin and eosin, Weigert's fuchselin stain for elastic tissue, and Weigert's hæmatoxylin and Van Gieson's stain, 3 to 6 sections of each tissue being examined.

RESULTS.

1. *Changes in the kidneys.*

In rabbits in which one renal artery was constricted, the opposite kidney having been removed or destroyed, the remaining kidney was hypertrophied

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† Since this paper was written Goldblatt (J. exper. Med., 1938, 67, 809) has published extensive observations on acute arterial lesions in dogs with experimental hypertension. Our findings closely resemble his, but he considers renal insufficiency to play a part in producing the lesions.

and appeared normal histologically. In rabbits in which both renal arteries had been constricted, one kidney was small and the other enlarged. The smaller kidney, which had sometimes undergone partial infarction, always showed fibrosis and tubular atrophy ; most of the glomeruli were normal, but many showed acute or chronic destructive lesions. The larger kidney appeared histologically normal except in rabbit 116 where very occasional glomeruli were pale and swollen with loss of structural detail.

2. *Arterial changes.*

(a) *Arteriosclerosis.* It is known that so-called arteriosclerosis occurs spontaneously in the rabbit. Changes of this type were observed in one rabbit (116) and were found in the larger arteries of kidney, myocardium and lungs, the lesions consisting of intimal fibrosis with splitting and reduplication of the elastica interna ; the aorta showed atheromatous changes to the naked eye and histologically medial calcification was present. These lesions of the larger arteries are mentioned to avoid confusion with the acute lesions of the smaller arteries described below.

(b) *Acute arterial lesions.* The occurrence and distribution of acute arterial lesions in the tissues of 11 rabbits with hypertension and of 9 normal rabbits are summarised in the accompanying Table 1. The lesions were found in none of the tissues of the control animals. In the rabbits with hypertension the severity and extent of the lesions seemed to be closely related to the degree of hypertension. Thus the most intense lesions were found in rabbits 39, 48, 54 and 116, in which the blood pressure attained the level of 130 mm. Hg or more. No acute vascular lesions were found in 5 rabbits and in 4 of these the pressure did not significantly rise above 105 mm. Hg. When present, the arterial changes were most severe and most frequent in the intestine ; they were present in occasional vessels in the stomach, suprarenal, liver, myocardium and eye. No lesions were discovered in the skin or striated muscle ; nor were arterial lesions found in the kidney of which the renal artery had been constricted except in rabbit 54, in which both renal arteries had been constricted. In this rabbit occasional arteriolar necroses were found close to the areas of infarction in the smaller kidney.

The acute arterial lesions found in the rabbits with severe hypertension are of two kinds, namely acute necrosis and cellular intimal thickening. The most constant feature of acute necrosis is the appearance in the vessel walls of amorphous (so-called fibrinoid) material which stains purplish red with hæmatoxylin and eosin (Fig. 1). In the smallest arteries and arterioles the whole structure of the wall may be destroyed and replaced by this substance which frequently extends into the perivascular tissue (Fig. 2). In larger arteries fibrinoid necrosis usually affects both intima and media but may be found in the intima or media alone (Figs 3-5). When the intima is affected it becomes swollen with fibrinoid material which separates

the endothelium from the elastica interna with great stretching and fragmentation of the latter; the lumen becomes greatly narrowed and sometimes obliterated. When the media is involved, muscle nuclei are destroyed and fragmented and the cells largely replaced by fibrinoid material. Plasma cells, eosinophil leucocytes and occasionally polymorphonuclear leucocytes may form a " periarterial granuloma " partly replacing the adventitia and media (Fig. 4). In occasional vessels red blood corpuscles are found in the media. These changes produce great thickening of the arterial walls ; although in smaller vessels this produces narrowing of the lumen, in some arteries of large calibre the elastica interna becomes stretched and the vessel appears dilated.

The second type of lesion, cellular intimal thickening, was found together with the acute necrotic lesions in 4 rabbits, and was present in the intestine, liver and heart muscle. In the elastic tissue stain (Figs 6 and 7), the elastica interna is seen to be intact but apparently stretched. The media often appears normal, but between the elastica interna and the endothelium there is a zone of cellular connective tissue which may cause great narrowing of the lumen. The elastic stripe of the intima is often hyperplastic in these vessels, producing a ring of elastic tissue immediately external to the endothelium. It is possible that this cellular intimal thickening is produced by organisation of the acute fibrinoid necrosis described above.

The diameter of the vessels affected by these lesions ranged from 40 to 250 μ, but in view of the great thickening of the wall and dilatation of the lumen, it is probable that the corresponding vessels in normal rabbits are of smaller dimensions.

Details of post-mortem findings.

RABBIT 39.

Before operation the blood pressure varied from 77 to 84 mm. mercury. The right kidney was removed on 15.1.37 and the left renal artery constricted on 8.2.37. The blood pressure was 103 mm. Hg on 17.2.37, 120 mm. on 3.3.37, 134 on 2.4.37, 153 on 7.4.37, 161 on 10.5.37, and from then until death on 9.6.37 remained over 140 mm. Hg.

Autopsy. Numerous petecheal hæmorrhages throughout the length of the gut, and in the myocardium ; hæmopericardium ; large retroperitoneal hæmorrhage extending from left kidney to left common iliac vessels. Left kidney enlarged but otherwise normal in appearance ; right kidney absent. Heart enlarged, particularly left ventricle. Aorta normal.

Microscopic findings.

Left kidney. Glomeruli appear normal; tubules show no changes apart from albuminous degeneration which is considerable in some areas ; no increase in interstitial tissue ; vessels quite prominent in intermediate zone, but no arterial lesions detected. *Small intestine.* Very numerous small arteries and arterioles in submucosa show acute fibrinoid necrosis with great swelling of their walls, narrowing of the lumen and exudation of fibrinoid material into the surrounding tissue. In the small arteries, the typical lesion is acute fibrinoid necrosis of the media without any intimal change. *Suprarenals.* Focal hæmorrhages in cortex. Fibrinoid necrosis of occasional arterioles. *Myocardium.* Focal interstitial hæmorrhage. Fibrinoid necrosis of several arterioles with extension of fibrinoid material into surrounding tissue. *Stomach, liver, striated muscle, skin,* nothing abnormal. *Spleen :* hyalinisation of many arterioles similar to changes present in spleens of normal rabbits.

Summary. In this animal with a severe degree of hypertension acute fibrinoid necrosis, chiefly affecting the media, was found in most of the arterioles and small arteries of the small intestine and in a few arterioles in the suprarenals and myocardium.

RABBIT 48.

Before operation the blood pressure ranged from 74 to 83 mm. Hg. The right ureter was tied on the 26.1.37 and the left renal artery constricted on 15.2.37. The blood pressure increased to 92 mm. Hg on 17.2.37 and thereafter rose gradually to 106 on 1.3.37, 130 on 9.4.37 and 146 on 27.4.37. The animal died on 28.4.37.

Autopsy. Numerous subcutaneous and intramuscular hæmorrhages, some very large. Numerous small hæmorrhages in large intestine, particularly terminal part where the lumen contains much blood. Heart enlarged, especially left ventricle. Right kidney a hydronephrotic sac. Left slightly enlarged but normal in appearance. Aorta normal.

Microscopic findings.

Left kidney. Histologically normal except for slight albuminous degeneration of the tubular epithelium. *Large intestine.* Very numerous small arteries in submucosa show acute fibrinoid necrosis. The vessels are dilated and their walls thickened. The elastic stain shows stretching and fragmentation of the elastica interna with deposition of amorphous material (taking the purplish stain of fibrin with hæmatoxylin and eosin) between elastica and endothelium. Necrosis of the media with deposition of fibrinoid material and red blood cells between the muscle nuclei. One portion of large intestine shows extensive hæmorrhage into the submucosa and mucosa. *Small intestine.* Very similar lesions in arterioles of submucosa but rather less frequent. *Stomach.* Several small arterioles show fibrinoid necrosis of their walls with obliteration of the lumen. *Myocardium.* Two arterioles show acute fibrinoid necrosis. One small artery shows conspicuous cellular intimal swelling inside a greatly stretched and fragmented elastica interna. *Retina.* Several arterioles show acute fibrinoid necrosis with great swelling of the wall. Red blood cells present within the wall of one arteriole. *Liver.* Several arterioles show cellular intimal thickening between elastica and endothelium. *Suprarenal, striated muscle, skin, ovary, spleen.* No acute arterial lesions detected.

Summary. In this animal with a severe hypertension acute arterial lesions were found in small and large intestine, stomach, liver, myocardium and retina. Both acute fibrinoid necrosis and cellular intimal thickening were present.

RABBIT 54.

Before operation the blood pressure averaged 67 mm. Hg. The right renal artery was constricted on 30.7.37 and the left on 11.8.37. The blood pressure rose to 82 mm. Hg on 12.8.37, 102 on 9.9.37, 129 on 21.9.37, and 137 mm. Hg on 9.10.37. On 11.10.37 the animal was killed.

Autopsy. Enlargement of the heart, particularly of the left ventricle ; a small scarred right kidney and an enlarged but otherwise normal left kidney. No hæmorrhages seen.

Microscopic findings.

Left kidney. Histologically normal except for albuminous degeneration of tubular epithelium. *Right kidney.* Reduced in size ; diffuse, moderately cellular fibrosis ; several areas of old infarction in cortex with complete destruction of renal elements ; elsewhere tubules atrophied, glomeruli reduced in number and mostly normal in appearance except adjoining areas of infarction, where occasional glomeruli show acute and chronic destructive lesions. The acute glomerular lesions very occasionally associated with acute necrosis of afferent arterioles. Otherwise arteries appear normal. *Large intestine.* Numerous small arteries show acute fibrinoid necrosis. Others show cellular intimal thickening with hypertrophy of elastic stripe of intima. *Small intestine.* Very numerous small arteries and arterioles show acute lesions similar to those in large intestine. *Liver.* One group of small arteries show cellular intimal thickening internal to the intact elastica interna. *Suprarenal.* One arteriole in medulla shows fibrinoid necrosis. *Skin, voluntary muscle, brain, stomach, spleen.* No acute vascular lesions seen.

Summary. In this rabbit with a severe hypertension acute arterial lesions were found in small and large intestine, liver and suprarenal. Two types of lesion were present, acute fibrinoid necrosis and cellular thickening of the intima.

RABBIT 116.

The arterial pressure before operation varied from 72 to 78 mm. Hg. On 17.8.37 the right renal artery was constricted without increase in the arterial pressure. On 20.9.37 the left renal artery was constricted. The arterial pressure rose to 100 mm. Hg on 2.12.37 and to 128 on 21.1.38, remaining between 120 and 133 until 23.2.38 when the animal was killed.

Autopsy. Left kidney small and calcareous ; right kidney enlarged but otherwise normal. Heart hypertrophied. Aorta thick and inelastic, atheromatous ulceration present.

Microscopic findings.

Right kidney. Intimal fibrosis with elastic reduplication in larger arteries (elastosis). *Left kidney.* Diffuse fibrosis with great tubular atrophy. Many glomeruli normal, others show all stages from acute fibrinoid necrosis to complete hyalinisation. A few larger vessels show elastosis. *Small intestine.* Many small arteries show advanced cellular intimal thickening with obliteration of lumen. *Large intestine.* Both cellular intimal thickening and acute fibrinoid necrosis present, most small arteries being affected. *Stomach.* Similar acute arterial changes to those in intestine but less severe. *Myocardium.* Intimal fibrosis of medium sized arteries with reduplication and splitting of elastica interna (elastosis). *Lung.* Pronounced elastosis of large and medium sized arteries. *Aorta.* Atheromatous changes with medial calcification *Liver, suprarenal, thyroid, eye.* No acute arterial lesions seen.

Summary. In this rabbit with moderately severe hypertension acute arterial lesions were present in small and large intestine and stomach. Both acute necrosis and cellular intimal thickening were present.

RABBIT 13S.

Before operation the arterial pressure varied from 75 to 79 mm. Hg. The right renal artery was clamped on 27.11.37 and the left on 1.12.37. The arterial pressure was 84 mm. Hg on 8.12.37, 102 mm. on 21.12.37, 123 mm. on 21.1.38 and remained between 114 and 134 mm. until the animal was killed on 4.4.38.

Autopsy. Simple atrophy of left kidney and enlargement of right kidney. Heart hypertrophied.

Microscopic findings.

Right kidney. No histological abnormality. *Left kidney.* Diffuse fibrosis and tubular atrophy. · Glomeruli crowded together in cortex but normal in appearance. No vascular lesions seen. *Suprarenals.* Acute necrosis in a small number of cortical arterioles. *Liver.* Cellular intimal thickening of occasional hepatic arterioles. *Small and large intestine, stomach, eye, spleen, skin, myocardium and skeletal muscle.* No vascular lesions seen.

Summary. In this rabbit with a moderate degree of hypertension occasional acute arterial lesions were present in the suprarenal and liver. Both acute necrosis and cellular intimal thickening were present.

RABBIT 38.

Arterial pressure before operation 64 to 75 mm. Hg. On 16.1.37, the right renal artery was ligated; on 8.2.37 the left renal artery was constricted. The arterial pressure rose to 84 mm. Hg. on 17.2.37, 109 mm. on 7.4.37 and remained between 85 and 108 mm. until 5.7.37 when the left renal artery was again constricted. The arterial pressure rose to 112 mm. on 15.7.37 and remained between 103 and 118 mm. until the animal was killed on 26.7.37.

Autopsy. Right kidney, a calcareous mass. Left kidney enlarged but otherwise normal in appearance. Heart hypertrophied. Aorta normal.

Microscopic Findings.

Small intestine. Several arteries show intense necrosis of media, with periarterial cellular reaction and very cellular thickening of intima with gross narrowing of lumen. *Large intestine, liver, suprarenal, myocardium and retina.* No arterial lesions found.

Summary. In this rabbit with a moderate degree of hypertension, acute arterial lesions of considerable severity were found in the small intestine only.

RABBIT 22.

Before operation the arterial pressure varied from 72 to 90 mm. Hg. After clamping the left renal artery on 23.5.36 the blood pressure varied from 72 to 92 mm. and on 22.6.36 the right renal artery was clamped. The arterial pressure remained unchanged for 3 months but on 3.11.36 it had risen to 105 mm.. The highest arterial pressure, 144 mm. Hg, was recorded on 6.4.37. From this time until the animal was killed on 4.4.38 the arterial pressure was between 120 and 135 mm. Hg.

Autopsy. Left kidney atrophied, right enlarged. Heart hypertrophied.

Microscopic findings.

Left kidney. Very dense fibrosis with tubular dilatation. Glomeruli crowded together but histologically normal. *Right kidney.* No histological abnormality. *Small and large intestine, stomach, spleen, pancreas, suprarenal, ovary, myocardium, skeletal muscle, skin and subcutaneous tissue, retina.* No vascular lesions found.

RABBIT 44.

The arterial pressure before operation averaged 78 mm. Hg. The right renal artery was ligated on 26.1.37 and the left constricted on 15.2.37. The arterial pressure rose to 94 mm. on 17.2.37 and 111 mm. on 22.3.37 and remained between 95 and 120 until 14.6.37 when the left renal artery was again constricted. The arterial pressure remained between 90 and 100 until the animal was killed on 3.8.37.

Autopsy. Right kidney very small and calcareous. Left kidney enlarged. Heart hypertrophied.

Microscopic findings.

The following tissues were examined :—

Small and large intestine, stomach, retina and aorta. No acute arterial lesions were found.

RABBIT 126.

The arterial pressure before operation varied from 82 to 88 mm. Hg. On 17.9.37 the right renal artery was clamped, the arterial pressure thereafter varying between 88 and 102 mm. Hg. The left renal artery was clamped on 1.11.37 and from 3.11.37 till 23.3.38, when the animal was killed, the arterial pressure lay between 100 and 115 mm. Hg.

Autopsy. Right kidney atrophied. Left kidney enlarged. Heart hypertrophied. Aorta normal.

Microscopic findings.

Right kidney. Diffuse fibrosis, large area of old infarction, glomeruli otherwise normal; no vascular lesions seen. *Left kidney.* No histological abnormality. *Large and small intestine, stomach, liver, spleen, pancreas, myocardium, skeletal muscle.* No vascular lesions found.

RABBIT 41.

The arterial pressure averaged 75 mm. Hg before operation. Right kidney removed on 16.1.37, left renal artery constricted on 8.2.37. On 23.2.37 the arterial pressure was 94 mm. and thereafter remained between 94 and 104 mm.. The animal was killed on 2.8.37.

Microscopic findings.

Small and large intestine, suprarenal, myocardium, retina, and aorta examined. No acute arterial lesions found.

RABBIT 115.

Before operation the arterial pressure ranged from 62 to 68 mm. Hg. The right renal artery was constricted on 17.8.37 and the left on 30.8.37. The arterial pressure rose to 80 mm. Hg on 1.9.37, 90 on 8.9.37, and from 11.10.37 until death on 10.1.38 remained between 92 and 114 mm. Hg.

Autopsy. Animal very thin. Numerous tubercles scattered throughout large and small intestine, particularly in appendix. Left kidney very small and largely calcareous. Right kidney slightly enlarged. Heart enlarged, particularly left ventricle.

Microscopic findings.

Right kidney. Histologically normal. *Left kidney.* Extensive old infarction; diffuse fibrosis in remnant of kidney tissue ; severe tubular atrophy ; glomeruli generally shrunken but appear otherwise normal.

No acute vascular lesions were discovered in *stomach, intestine, pancreas, suprarenals, liver, spleen, myocardium, or thyroid.*

DISCUSSION.

The association of hypertension in man with changes in the arteries and arterioles has long been recognised. Jores (3) first gave a clear account of the nature and distribution of arteriolo-sclerosis in the condition now recognised as benign hypertension. The characteristic lesion is fatty hyaline degeneration of the arterioles which is most marked in the vasa-afferentia of the kidney, but is also found in other abdominal viscera, especially pancreas, suprarenal, and intestine. It is generally believed that

these changes in the arterioles are the result and not the cause of the hypertension. In addition to fatty hyaline degeneration of the arterioles, the larger arteries in all cases of high blood pressure show some degree of "elastosis." This consists of thickening of the intima with proliferation of fibrous and elastic tissue and reduplication of the internal elastic lamina. This change is well seen in the interlobular and arcuate arteries of the kidney. It is not peculiar to hypertensive subjects but develops in the majority of individuals as age advances. Its association with hypertension is usually regarded as an acceleration of the ageing process. It must be emphasised that fatty hyaline degeneration of the arterioles and elastosis of the larger vessels are to be found in all cases where longstanding hypertension has been present during life, that is, in benign hypertension, malignant hypertension, and chronic nephritis.

In 1919 Fahr (1) described *acute* arterial lesions which were invariably present in the condition termed by him malignant nephrosclerosis. The lesions were of two types, the first an acute necrosis of the vessel wall with fragmentation of nuclei, deposition of fibrinoid material, and the occasional presence of red blood cells in the wall ; the second a cellular thickening of the intima without splitting or reduplication of the elastica interna. Fahr pointed out that these lesions were most severe and widespread in the kidneys, and occurred less constantly in other organs, particularly in the pancreas, suprarenals, and intestine. These lesions are not confined to malignant nephrosclerosis ; thus Löhlein (4) described acute necrotic lesions of the renal arterioles in severe cases of subacute diffuse nephritis. Furthermore an extensive study of the vascular lesions in Bright's disease by one of the present authors (C.W.) has revealed both types of arterial lesion in a number of cases of chronic diffuse nephritis, not only in the kidney, but also in the pancreas, suprarenals, intestine and retina. The subjects presenting these lesions had during life a severe hypertension and in the terminal stages the clinical picture closely resembled that of malignant hypertension.

The acute arterial lesions we have described in rabbits with experimental hypertension seem to be structurally identical with the lesions described by Fahr (Fig. 8). Their distribution in the rabbit and in man is also similar, with one outstanding exception, for in the rabbit these lesions are absent from the kidney, the organ which in man is most constantly and most severely affected. In the rabbit, however, the renal artery has been constricted and it is natural to suspect that the sparing of the kidney may be due to this, a possibility which is being tested experimentally.

In man, the presence of acute arterial lesions in malignant hypertension and in chronic diffuse nephritis when the arterial pressure is very high, together with their absence in benign hypertension where the diastolic pressure tends to be lower, suggest that the production of the lesions depends on the height of the blood pressure. Against this suggestion it may be argued that in man, some factor other than hypertension may cause the acute

arterial necrosis. Fahr for example, considers that necrotising arteriolitis is the primary lesion in malignant sclerosis, and gives rise to hypertension by narrowing the renal vascular bed. In the rabbit the problem is simplified for we have animals with different degrees of hypertension produced by deliberate interference with the renal blood flow. In such animals acute arterial lesions cannot be primary, and, as we have seen, their appearance

I. *Distribution of acute arterial lesions in rabbits with experimental renal hypertension.*

Rabbit.	Initial B.P.	Final B.P.	Duration of hypertension.	Large Intestine.	Small Intestine.	Stomach.	Adrenal.	Liver.	Myocardium.	Retina.
39	82	145	4 months		+	—	+	—	+	
48	77	135	2 ,,	+	+	+	—	+	+	+
54	70	135	2 ,,	+	+	—	+	—	—	
116	75	130	5½ ,,	+	+	+	—	—	—	—
138	77	125	4 ,,	—	—	—	+	+		
22	80	125	17 ,,	—	—	—	—		—	—
38	66	115	5 ,,	—	+	—	—	—	—	—
44	78	105	5½ ,,	—	—	—				—
126	86	105	5 ,,	—	—	—			—	
41	74	100	5 ,,	—	—	—	—		—	—
115	66	100	3 ,,	—	—	—	—	—	—	—
Controls. (9)	65-85			—	—	—	—	—	—	—

     +   acute arterial lesions present.
     —   ,,   ,,   ,,   not found.

     The following organs were examined and showed no acute arterial lesions :—kidney, ovary, skeletal muscle, skin, thyroid, brain, spleen, pancreas.

seems to be related only to one factor, the presence of a rapidly developing severe hypertension; in animals with a moderate hypertension of long duration, acute lesions are not found. In view of the lack of demonstrable structural change in the kidneys it appears that renal failure plays no part in the process. In agreement with the view that the lesions are the result

of high blood pressure is their absence from the kidney, where it may be inferred the arterial pressure is less high owing to the clamp on the renal artery.*

This view which we have put forward of the pathogenesis of acute arterial lesions, appears to be that which fits the known facts and one which seems to be open to experimental verification.

SUMMARY.

1. Acute arterial lesions structurally identical with those of malignant hypertension in man have been found in rabbits with arterial hypertension produced by renal artery constriction. The incidence of the lesions was related to the degree of hypertension but not to its duration.

2. These lesions were most frequnt and severe in the intestine, but were also found in stomach, liver, suprarenal, heart, and eye ; they were absent from the kidney, the renal artery to which had been constricted.

3. It is suggested that a greatly raised intra-arterial pressure is a chief factor determining these lesions in human and experimental hypertension.

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* The pressure in the artery distal to the clamp has not been measured in rabbits ; but in the dog Blalock and Levy (Ann. Surg., 1937, 106, 826) found it considerably below the systemic arterial pressure.

Fig. 1.

Fig. 2.

Fig. 3.

Fig. 4.

Fig. 1. (× 330). Rabbit 54. Large intestine. H and E stain. Fibrinoid necrosis with obliteration of lumen.

Fig. 2. (× 140). Rabbit 39. Myocardium. H and E stain. Fibrinoid necrosis of arterioles with extension of fibrinoid substance into perivascular tissue.

Fig. 3. (× 220). Rabbit 54. Same vessel as in Fig. 1. Elastic tissue stain. Fibrinoid necrosis confined to intima. Great stretching of elastica interna.

Fig. 4. (× 330). Rabbit 48. Large intestine. Elastic tissue stain. Fibrinoid necrosis of small artery affecting both intima and media.

EXPERIMENTAL HYPERTENSION OF RENAL ORIGIN IN THE RABBIT.*

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THE production of hypertension in the experimental animal by interfering with its kidneys is of interest to medicine chiefly because raised arterial pressure in man is believed to be also of renal origin at least in certain instances ; it is hoped that a solution of its mechanism in the animal may accelerate the solution in man. The simplest and most certain method of producing hypertension of renal origin seems to be that of constricting the renal arteries by silver clamps as described by Goldblatt and his co-workers (8). The method has been extensively used in dogs and also in monkeys (5), but there are many workers, particularly in this country, who are forced to work on smaller animals ; of such the rabbit is one of the most suitable because of the ease with which its blood pressure can be estimated. The present paper describes a simple modification of Goldblatt's method for constricting the renal arteries, and some of the resultant changes in the rabbit. Results obtained on other species by previous workers using similar methods have recently been summarised by Goldblatt (6), and will not be fully referred to here.

Method.

We used brown adult female rabbits fed on a liberal diet of bran, oats, and green vegetables. Blood pressures were measured by Grant and Rothschild's capsule (10) on the central artery of the ear. Measurements were made in a warm room (about 24°C) into which the animals were brought half an hour or more previously. Each animal was put on a comfortably warm pad, and covered with a blanket secured at the sides with sandbags. In these circumstances, the rabbits would remain quiet with their ears flushed. The blood pressure was recorded at intervals of about 30 sec. until it ceased to show a progressive change ; four successive readings were

averaged. Readings of arterial pressure obtained in this way agree closely with the mean pressure recorded by cannulation of the femoral artery (10, 16).

A simple clamp for constricting the renal arteries may be made as follows (Fig. 1). From a strip of standard silver, 0·5 mm. thick and 6 mm. broad, a piece 14 cm. long is cut ; the edges are rounded with a file. The strip is then bent into the shape of a U. A metal plate of known thickness (usually 0·5 mm.) is passed between the limbs of the U which is then firmly compressed in a vice. After releasing the compression the U shaped strip maintains its shape fairly well, the limbs remaining apart by a distance just greater than the thickness of the plate used to separate them. The internal diameter of the clamp is measured accurately with a microscope and micrometer eyepiece. By using plates of varying thickness, clamps of varying size may be made.
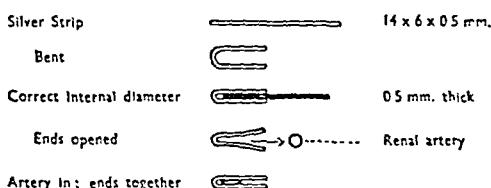
Fig. 1. A diagram to illustrate the clamp for constricting the renal artery. The first three stages represent the construction of the clamp and the adjustment of its internal diameter. The last two stages illustrate its application to the renal artery.

The clamp is applied to the renal artery as follows. Operating with aseptic precautions under ether or nembutal anæsthesia the kidney is exposed through the loin, mobilised by freeing it from surrounding connective tissue and delivered through the wound. The renal artery and vein are cleaned from adherent fat and separated from one another for a suitable distance by a flat blunt probe. The free ends of the two limbs of the clamp are separated by the handle of a scalpel, the clamp is slipped on to the artery, and the free ends approximated by squeezing them in a pair of artery forceps. The clamp usually returns to its original shape, and although its free ends remain a little separated, it rarely slips off the artery subsequently. The kidney is then returned to the abdomen and the wound sown up. The right kidney lies partly under the ribs, and its exposure is facilitated by a sand bag the centre of which is placed under the left costal margin, the animal lying on its left side. The left renal artery is long, and if the first clamp constricts too little, a second and even a third may be applied proximally by exposing the renal artery through an anterior abdominal incision. The right renal artery is short, and can usually be clamped only once, and most easily through the loin.

The only difficulty in using the clamps is in adjusting their size to obtain the desired result. Clamps less than 0·4 mm. internal diameter usually cause necrosis of the kidney and when wider than 0·7 mm. may not

cause any hypertension. The most useful size for producing persistent hypertension is about 0·5 to 0·6 mm., but with such clamps there are failures either from necrosis of the kidney or from inadequate constriction.

As far as we can tell the only action which the clamps can exert is to reduce the pressure in the renal artery as it enters the kidney. The clamps are applied close to the hilum of the kidney and the only branches arising from the renal artery distal to the clamp are those to the kidney and a small branch anastomosing with the ureteric artery.

Results.

Ligature of the ureter. Four rabbits were observed for two to four weeks after ligating one ureter, the other kidney remaining intact; no definite rise of blood pressure occurred; in all instances a hydronephrosis was found at autopsy.

Ligature of the renal artery. Ligature of one renal artery, the other kidney being intact, produced no rise of blood pressure in 9 rabbits observed for 2 to 20 weeks. The procedure was followed by necrosis of the kidney


Fig. 2. The arterial pressure of rabbit 114 before and after applying a 0·5 mm. clamp to the right renal artery on the 8th day. At autopsy 15 weeks after operation, the right kidney was very small (1·8 g.) but of normal consistency apart from a calcareous scar at one corner.

and its ultimate shrinkage to a small yellow calcareous lump weighing about 1 g.. The bloodflow through the kidney is probably not abolished entirely by ligating the artery, because a few days after this operation a lead chromate gelatin mass injected into the abdominal aorta at a pressure of about 100 mm.Hg, enters some of the interlobular and afferent glomerular arteries of the necrotic kidney by way of anastomotic arteries passing up the ureter to the hilum of the kidney. There exists therefore a possible channel through which a small amount of blood can flow to the necrotic kidney, and through which the products of renal necrosis can enter the blood stream. The absence of any definite rise of blood pressure is against such necrotic products being the cause of renal hypertension, as has been suggested in the past, a conclusion with which subsequent observations agree.

Constriction of one renal artery, the other kidney being intact. In 17 animals, clamps varying from 0·5 to 0·7 mm. in internal diameter were

applied to the right renal artery, the left kidney being intact. A rise of blood pressure of 10 mm. or more above the pre-operative level occurred in 5 animals, but in only one was the rise as much as 30 mm. Hg and in this transient (Fig. 2) ; in this animal the right kidney was found at autopsy to
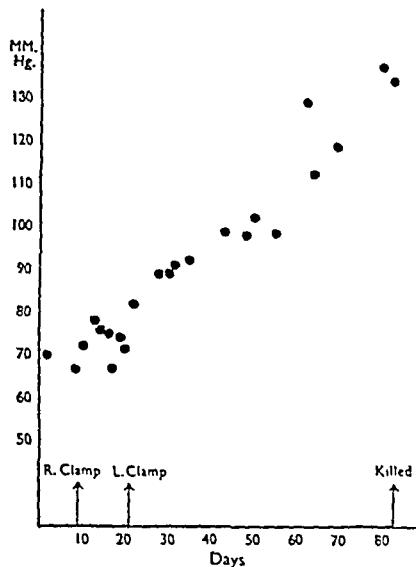

Fig. 3. Rabbit 54. Constriction of the right, and subsequently the left, renal arteries. At autopsy the right kidney was small and partly calcified ; the left was enlarged but of normal appearance.
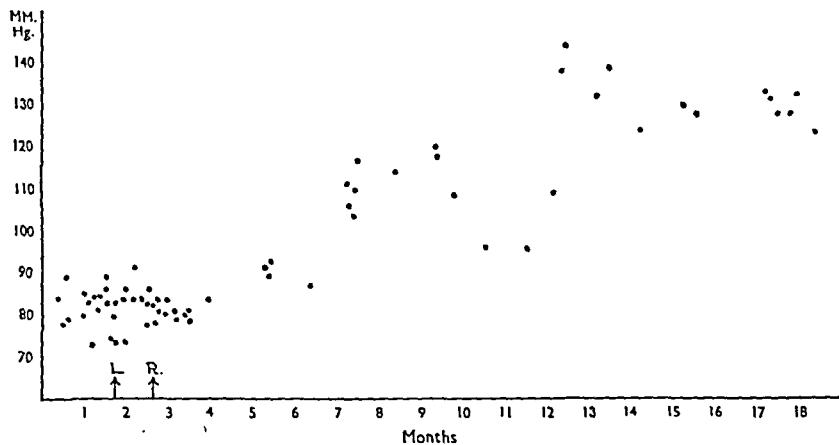
Fig. 4. Rabbit 22. Constriction of the left, and 1 month later of the right, renal arteries. This animal was killed 22 months after the first operation, its blood pressure being between 120 and 130 mm. Hg. Autopsy showed simply atrophy of the left and enlargement of the right kidney.

have atrophied very considerably, weighing only 1·8 g. as against 13·7 g. for its fellow. A simple atrophy of the ischæmic kidney was also observed in one animal in which no hypertension developed, and one in which it was

slight. Atrophy of the ischæmic kidney with enlargement of the other suggests that when the blood supply to one kidney is reduced, the work previously done by it may be transferred to its fellow ; such a kidney, while suffering a reduction in its blood supply would probably have also a reduction in its blood demand. The absence of any pronounced hypertension in these circumstances suggests that the stimulus to the rise of arterial pressures may be an upset in the normal relationship of blood supply and demand in the kidney, rather than a simple ischæmia.

Constriction of both renal arteries. In 15 rabbits the left renal artery was constricted 1 to 4 weeks after the right, the blood pressure at the time of the second operation being normal or but slightly raised. The internal diameter of the clamps used was 0·5 mm. to 0·7 mm.. Three animals died within 3 days of the second operation and at autopsy showed necrosis of the left and atrophy of the right kidney. Persistent hypertension developed in 9 and was moderate or severe in 5. Fig. 3 exemplifies a severe hypertension. Fig. 4 illustrates an experiment which was remarkable for the long interval which elapsed before the blood pressure began to rise.
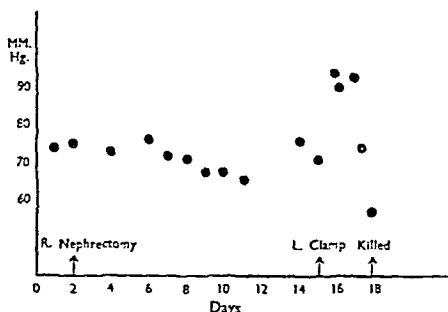

Fig. 5. Rabbit 45a. The right kidney was removed on the 2nd day, and a 0·37 mm. clamp applied to the left renal artery on the 15th day. At autopsy late on the 17th day the kidney was found enlarged and soft, and showed patches of necrosis.

In 5 animals that have come to autopsy the clamps have been unequal in size and the kidney having the narrower clamp has been atrophied or partially or completely infarcted, the other kidney being enlarged but normal macroscopically. It is possible that these changes in the relative sizes of the kidneys may be in part responsible for the onset of hypertension being so greatly delayed as in Fig. 4.

In its essence, therefore, this method of producing hypertension resembles the next in that it seems to depend on destroying one kidney and producing adequate ischæmia of the other. But it is less satisfactory in being less deliberate.

Constriction of the renal artery to the only functioning kidney. The most satisfactory method of producing renal hypertension in the rabbit is to remove
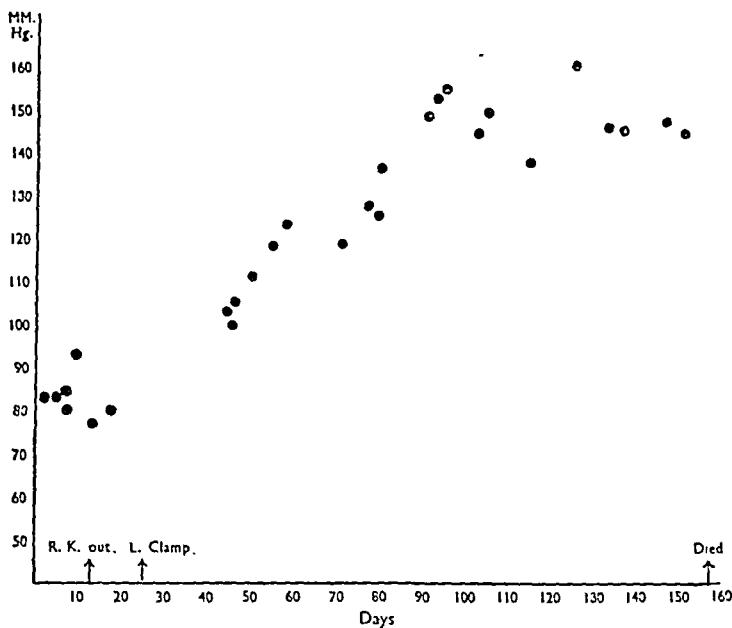
Fig. 6. Rabbit 39. The right kidney was removed and the left renal artery constricted with a 0·6 mm. clamp. At autopsy the animal showed multiple small and large hæmorrhages in gut, muscle and heart. The left kidney was enlarged but was otherwise normal.
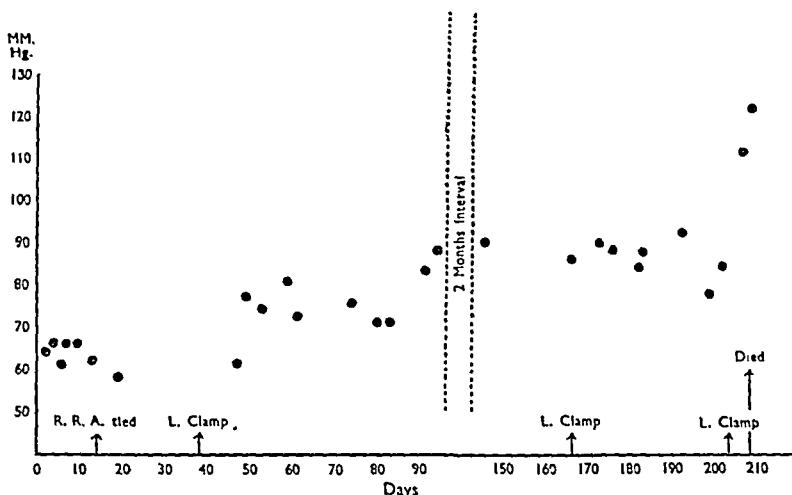

Fig. 7. Rabbit 40. Ligature of the right renal artery, constriction of the left. Two months during which the hypertension was unchanged are omitted from the record. A second 0·7 mm. clamp was applied to the left renal artery on the 166th day without producing any further rise of blood pressure ; the application of a third and narrower clamp on the 203rd day led to an acute rise of blood pressure and death on the 208th day. At autopsy the left kidney was found greatly swollen with areas of necrotic cells and neighbouring areas of inflammation.

the right kidney, or to destroy it by tying its artery or ureter, and then after 2 weeks or longer to constrict the left renal artery. The effects observed are chiefly dependent on the degree of constriction employed.

With clamps having an internal diameter of about 0·35 to 0·4 mm. the blood pressure is raised by 10 to 30 mm. within 6 hours after operation and remains raised until late on the 2nd day when it falls to normal or below, and the animal dies (Fig. 5). At autopsy, 50 to 60 hours after operation, the kidney is enlarged and soft and presents pale and hæmorrhagic areas of varying extent. The ureter and bladder contain a little red urine in which are numerous red cells and cellular casts. Sections of the kidney show that in the pale areas the kidney cells are necrotic and the tubules and glomeruli are distended with an albuminous content; in the intervening red areas, the renal cells seem intact but there is intense vascular engorgement and infiltration of inflammatory cells. In such kidneys it is evident that the ischæmia has been severe enough to produce some renal necrosis. Yet the clamp does not actually occlude the renal artery, for methylene blue injected at a pressure of 20 mm. Hg into the aorta will flow through the renal artery past the clamp.

With slightly larger clamps, 0·5 to 0·7 mm. in internal diameter, the arterial pressure rises more slowly but the kidney and animal survive and a lasting hypertension may be obtained. Because the rise of blood pressure is slow, it is difficult to be sure when it begins; it may be definite as early as the first or as late as the 10th day and afterwards continues to increase for 1 to 3 months, when the pressure may become more or less stable. An example of severe hypertension produced in this way is given in Fig. 6 and one of a mild hypertension in Fig. 7. Fig. 7 also shows that when clamping the renal artery leads only to a moderate hypertension, increase in the ischæmia may be followed by a further rise in blood pressure.

Remarks on rabbits with hypertension.

Rabbits with persistent hypertension usually appear undistinguishable from normal rabbits. They maintain their weight unless afflicted with intercurrent disease. They may beget, deliver and suckle young. The following points require special mention.

Urine. In rabbits developing acute hypertension as a consequence of severe ischæmia and partial necrosis of the only kidney, very little urine is formed and this contains much blood, protein and cellular casts. Hæmaturia also occurs very commonly for a day or two after any operation in which the kidney is handled. Several weeks after operation animals with hypertension commonly show no red cells or casts in the urine.

Blood urea. The blood urea lay between 30 and 35 mg. per 100 c.c. in 3 animals with blood pressures of 115 to 130 mm. Hg 2 to 4 months after constricting both renal arteries. Goldblatt and his colleague (8) have shown that in the dog hypertension may occur with a normal renal functions

Vascular reactions. The ears flush and pale as the environment is warm or cool ; when flushed the ears appear normally so.

Grant (9) has shown that in the rabbit the vessels of a sympathectomised ear regain tone, being less dilated than the vessels of a normal ear when the animal is warm ; the regain of tone is associated with a heightened reactivity to many vaso-active substances, though probably not to all. It seemed possible that if renal hypertension were due to a chemical agent, this might act more strongly on sympathectomised than on normal vessels, and that the difference between sympathectomised and normal ears might be accentuated with the onset of hypertension. In two white rabbits, the left ear was sympathectomised by cutting the great and posterior auricular nerves and by removing the superior cervical sympathetic ganglion on that side. A month or more later, operations on the kidneys were begun ; in one animal the right kidney was removed and the left renal artery constricted ; in the other the two renal arteries were constricted. With the animals warm and quiet, the temperature of the ears was recorded thermo-electrically from symmetrical points and the ears were photographed ; this was done repeatedly before operating on the kidneys and also 4 to 6 months afterwards when the blood pressures were between 110 and 130 mm. Hg. Before the renal operations the sympathectomised ear was a little paler and cooler (0·5 to 2·0°C.) than its fellow when this was flushed. After the development of hypertension this difference remained unchanged. The experiment suggests that if this form of hypertension is chemical in origin, then the chemical substance is not one to which the sympathectomised vessels are unusually susceptible.

According to Koch, Mies and Nordmann (14), rabbits in which hypertension has been produced by cutting the carotid sinus and aortic nerves, respond to a needle prick with a fall of blood pressure. Our rabbits with hypertension have responded to a prick with a rise of blood pressure, as do normal animals.

The heart. In the rabbits with hypertension and in a series of 32 normal rabbits the heart was cut out by severing the great vessels at their origin, stripped of its pericardium and adherent tissue, and washed free of blood. After removing adherent water by squeezing in filter paper the heart was weighed. The rabbit was also weighed before (total weight) and after (carcase weight) removing the abdominal viscera. In our series of normal rabbits we found heart weight related rather more closely to the carcase weight than to the total weight, a considerable factor in the latter being the contents of the gut. The relationship between carcase weight and the ratio $\frac{\text{heart weight} \times 100}{\text{carcase weight}}$ in the normal rabbits and in 16 rabbits with chronic hypertension is shown in Fig. 8. The hypertensive rabbits had had blood pressures of 100 mm. Hg or more for 1 to 17 months. It may be seen that 4 of the hypertensive heart weights fall within the upper limits of normal ; the remainder are heavier and sometimes much heavier than normal. Fig. 9
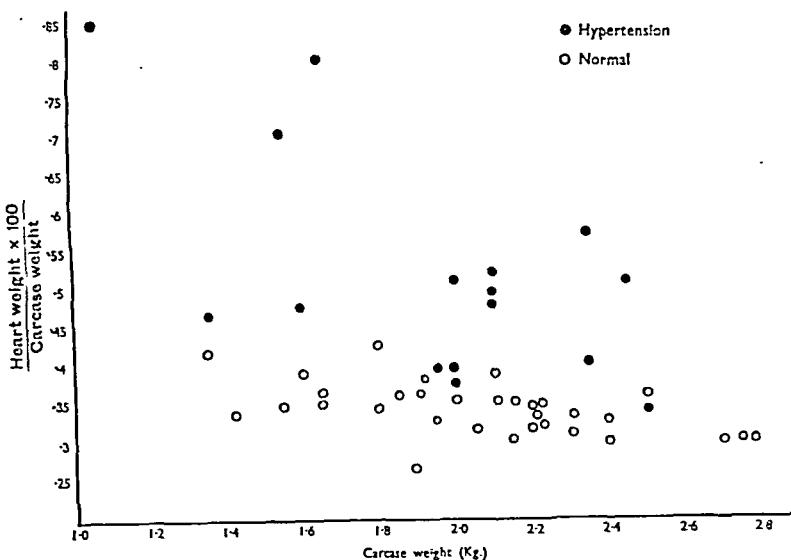
Fig. 8. The relationship between the weight of the eviscerated carcase and the ratio $\dfrac{\text{heart weight} \times 100}{\text{carcase weight}}$ in normal rabbits (open circles) and hypertensive rabbits (black discs).
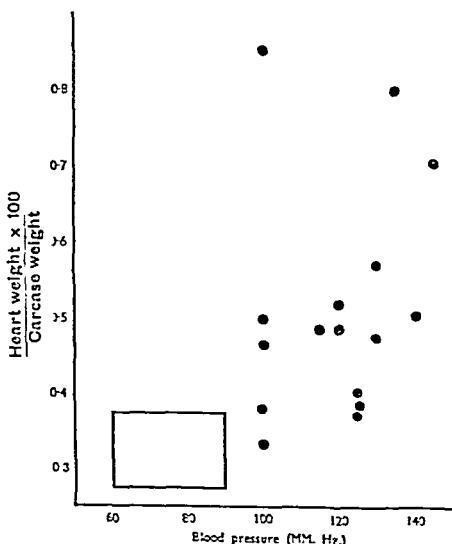

Fig. 9. The relationship between the blood pressure and the ratio $\dfrac{\text{heart weight} \times 100}{\text{carcase weight}}$ in rabbits with hypertension for 1 month or more. The rectangle represents the normal range.

shows the relationship between the arterial pressure before death and the heart-carcase weight ratio in the rabbits with chronic hypertension ; it will be seen that there is a tendency for the degree of cardiac hypertrophy to be proportional to the degree of hypertension, but the relationship is not close. The chief exception is the rabbit with the largest heart-carcase ratio and a blood pressure of only 100 mm. Hg ; the high ratio was due not to a particularly large heart but to an abnormally small carcase, the animal being much emaciated as a result of pseudo-tuberculosis of which it died. In other animals intercurrent disease was absent. We have not compared the weights of the separate chambers, but it is clear from naked eye inspection of the heart that hypertrophy in the hypertensive rabbit is chiefly of the left ventricle.

Hypertension resulting from renal artery constriction in the rabbit thus resembles persistent hypertension in man in its association with conspicuous cardiac hypertrophy. The occurrence of cardiac hypertrophy in the hypertensive dog with its renal arteries constricted is suggested by the figures of Collins (3) and Elaut (4), but in this animal heart weight and body weight are less constantly related than in the rabbit and cardiac hypertrophy correspondingly difficult to demonstrate. In rats with raised arterial pressure following partial nephrectomy, Chanutin and Ferris (2) have found an increase in heart weight which is related in degree to the elevation of the arterial pressure. On the other hand Hamperl and Heller (11) were unable to demonstrate cardiac hypertrophy in dogs with chronic hypertension produced by intracisternal injection of kaolin. In rabbits with prolonged hypertension following section of the carotid sinus and depressor nerves, Nordmann (15) and Boyd and McCullagh (1) have demonstrated an increase in the weight of the left ventricle relative to the right. Taking the ratio between heart weight and body weight as 0·24% in normal animals, Nordmann found increase of 11·4% to 30% in this ratio in 7 rabbits with arterial pressures of 145 to 171 mm. Hg. In our series of 32 normal rabbits, $\dfrac{\text{heart weight} \times 100}{\text{body weight}}$ had a mean value of 0·254 and limits of 0·20 and 0·33. The heart weight of Nordmann's hypertensive rabbits are thus all above our normal mean, but only two fall above our normal limits ; the degree of cardiac hypertrophy which he observed thus seems to have been much less than that found in our series. A possible reason for this difference is that the arterial pressure was in general higher in our series than in his, for we measured systolic pressure in the ear and in Nordmann's series the pressure seems to have been measured in the exposed carotid artery by the device of Koch and Mies (13). Moreover, according to Heymans and Bouckaert (12), the arterial pressure in dogs without carotid sinus and depressor nerves is elevated only when the animal is awake and excited ; when quiet or asleep the arterial pressure tends to be normal.

The aorta was normal to naked eye examination in all our hypertensive rabbits except one, with a hypertension of 130 mm. Hg of 5½

months duration, in which the aorta showed extensive calcification and ulceration.

Arterial lesions. Two or our rabbits with severe and progressive hypertension died with multiple hæmorrhages in the gut and other tissues. It occurred to us that these animals might show acute arterial lesions and their tissues and those of other animals were examined histologically. This histological examination, already published (18), showed acute lesions, closely resembling those of malignant hypertension of man, in the small arteries and arterioles of the gut and other organs, but not in the kidneys, of animals with severe hypertension ; facts suggesting a greatly raised intra-arterial pressure as the chief factor determining these lesions. Similar lesions have been described by Goldblatt (7) in dogs with hypertension following renal artery constriction. They have not been found in dogs with kaolin hypertension (11), nor in rabbits with hypertension following removal of the carotid sinus and depressor nerves (1, 15). The absence of acute arterial lesions in these forms of hypertension seems to be correlated with the absence, or relatively slight degree, of cardiac hypertrophy, and may prove susceptible of the same explanation, namely that the elevation of arterial pressure is smaller or less persistent than often happens after renal artery constriction.

Comment.

In these experiments we may suppose that hypertension has been a direct or indirect response to some change in the renal circulation consequent on clamping the renal artery. As to how this response is brought about we have no new evidence to offer. We do not propose to discuss the old, but we would like to point out two features of the response in the rabbit which also seem to occur in the dog and have not received the attention they merit. The first is the latent period which may elapse between renal artery constriction and the subsequent rise of blood pressure. At one time we considered that this latent period must represent the lag between the change in circulatory conditions in the kidney and the initiation of the factor responsible for hypertension. However, it now seems probable that in many instances the latent period may represent the time taken for the renal circulation to adjust itself after operation, for the latent period is most pronounced in animals in which both renal arteries are constricted, and in such it is usual for one kidney to atrophy and the other to hypertrophy, changes which must themselves take a considerable time to complete. The second feature of the response, which is found in all animals surviving more than a few days, is the slowly progressive nature of the rise of pressure, which often takes a month or more to reach completion. We could most easily explain this phenomenon by supposing that the change in the kidney producing hypertension is one of slow and gradual development, but we are aware that other progressive changes, such as cardiovascular hypertrophy and in some instance acute arteriolar lesions may contribute.

In a previous paper (12) it was pointed out that experimental hypertension produced by section of the carotid sinus and depressor nerves differed in most major characteristics from persistent hypertension in man. It is satisfactory to note that increasing knowledge serves to emphasise the similarity between human hypertension and experimental hypertension produced by renal artery constriction ; in this connection the occurrence of gross cardiac hypertrophy and acute arterial lesions in hypertensive rabbits needs to be emphasised.

SUMMARY.

1. A simple method is described of making clamps of known size suitable for constricting the renal artery in the rabbit.

2. In the rabbit, constriction of one renal artery, the other kidney being intact, is sometimes followed by a small and transient rise of blood pressure, and usually by atrophy of the ischæmic kidney.

3. Constriction, by suitable clamps, of both renal arteries or of the renal artery to the only functioning kidney is followed by a slow rise of arterial pressure to a level which may be nearly double the pre-operative value. In such animals with hypertension, the heart is hypertrophied and the degree of hypertrophy seems to be related, though not closely, to the degree of hypertension.

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OBSERVATIONS ON THE PITUITARY CONTROL OF CREATINE AND CREATININE EXCRETION.

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THE excretion of creatine and creatinine in acromegalic subjects was described and discussed by Schrire (1), who observed that there was considerable divergence from what is commonly regarded as the normal. The abnormality in the elimination of these two substances was ascribed to alterations in function of the anterior lobe of the pituitary gland.

In acromegalics, the elimination of both creatine and creatinine in abnormally large quantities was a conspicuous feature. With the aid of injections of thyreotropic extracts and another preparation of the anterior lobe (antuitrin), it was possible to produce in normal individuals changes in the excretion of creatine and creatinine comparable to those seen in acromegaly. Antuitrin (a crude preparation of the anterior lobe) increased the excretion of creatinine in the urine without any effect on the creatine output. Thyreotropic extracts of the pituitary produced a profound creatinuria but in no way altered the level of urinary creatinine.

It was accordingly considered advisable to investigate the identity and the mode of action of the pituitary principles responsible for the variations in the output of creatine and creatinine.

Nitzescu and Gontzea (2, 3) have recently reported results obtained on creatine-creatinine excretion in certain abnormal subjects following injection of a growth extract, and a urinary gonadotropic extract. The purpose of the present paper is to describe and discuss the results obtained in normal individuals following the use of various pituitary extracts, and the significance of these findings will be discussed in relation to acromegaly.

Methods and material.

Quantitative estimations of creatine and creatinine in the urine were determined by Folin's colorimetric method (4). The measures adopted for collection of the urine have been noted in a previous communication (1).

The subjects chosen for this investigation were almost without exception in hospital for treatment of gastric or duodenal ulcers. All the subjects were ambulant, and the creatine-creatinine excretion of each individual before an experiment was within normal limits. These individuals with a normal creatine-creatinine metabolism will in this paper be alluded to as "normal subjects."

As the diet during the period of the experiment was meat and fish free, this was tantamount to a creatine free intake of food.

The following preparations were used :—

Thyreotropic extract.

Ambinon, (Organon Labs.) ; 1 ml. of the extract contains 100-300 Heyl-Laqueur units. This preparation is extracted from the anterior lobe of the pituitary gland of hogs, and contains some gonadotropic principle which cannot be eliminated without diminishing the thyreotropic activity.

One guinea-pig unit of Heyl-Laqueur is half the daily dose that, injected intraperitoneally on two successive days, induces a given degree of thyroid epithelial development in at least 66 per cent. of the test animals within 48 hours of the first injection.

Gonadotropic extract.

A. *Extracted from the anterior pituitary.* A special preparation from Organon Labs.. It is manufactured from sheep pituitaries (which have a low thyreotropic content) and contains the ovary follicle stimulating principle. There is some thyreotropic principle present as an unavoidable contaminant.

B. *Extracted from the serum.* Antostab (Boots) ; it is extracted from the serum of pregnant mares, and contains an ovary follicle stimulating principle.

C. *Extracted from the urine.* 1. Pregnyl (Organon). Extracted from the urine of pregnant women. Contains the ovary luteinising principle. 2. Antuitrin S. (P. Davis). A preparation from the urine of pregnant women containing the luteinising principle.

Growth extract.

A. Organon Labs. preparation. This is prepared from the anterior pituitary, and tested free of gonadotropic and thyreotropic principles. It is standardised on the rate of growth of hypophysectomised rats.

B. Antuitrin G. (P. Davis). Extracted from the anterior lobe of the pituitary. It does not contain a significant amount of thyreotropic or gonadotropic principles.

All extracts were administered by the intra-muscular route.

RESULTS.

Injection of thyreotropic extracts in normal subjects.

The effects of this procedure were previously described (1) on four subjects. This has been repeated on eight more normal subjects, the results being essentially the same. Fig. 1 represents graphically the effects on creatine excretion in one of these individuals. The subject was a normal male adult. On May the 27th and on the two succeeding days he was injected with one millilitre (ml.) of the thyreotropic extract. The quantity

of creatinine excreted was unchanged, but creatine appeared in the urine in large quantities. Before and after the period of injection no creatine was detected in the urine except in the three days immediately after cessation of the injections.
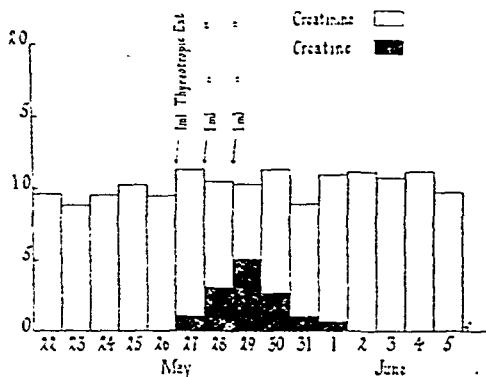

Fig. 1. Normal subject of 126 lbs. weight. Chart of urinary creatine and creatinine excretion in g. per 24 hours.
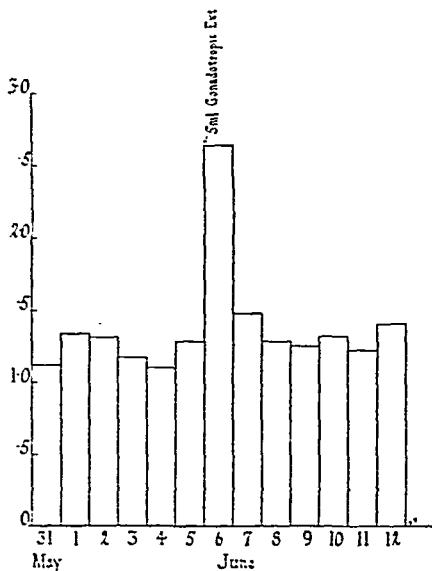

Fig. 2. Normal subject of 140 lbs. weight. Chart of urinary creatinine excretion in 24 g. per hours.

All twelve normal individuals showed the same results. Five abnormal subjects, investigated for other purposes, produced comparable results following the administration of thyreotropic extract.

Single large doses produced a marked transient increase of creatinine in the urine comparable to the output in acromegalic subjects. Injections of the follicle stimulating preparation from the serum (antostab), and the luteinising factor from pregnancy urine (pregnyl), (antuitrin S.) produced no alteration in creatine or creatinine elimination. Of the three preparations containing gonadotropic principle (pituitary, serum and urine), the only one to mobilise creatinine was a preparation derived from the pituitary gland. Creatine was unaffected by all three in single large injections. The principle derived from the pituitary is unavoidably contaminated with some thyreotropic principle, and in subjects injected with small doses of gonadotropic extract daily for a long period, creatine eventually appeared in the urine in small quantities due to the cumulative effect of the thyreotropic contaminant. In addition the basal metabolic rate was raised.

The administration of growth extract from the anterior lobe of the pituitary was without effect. The Organon preparation used was tested free of gonadotropic and thyreotropic principles, and had no effect on creatine or creatinine excretion. Antuitrin G. which has insignificant amounts of these two principles, similarly had no effect. Thus, of all the available principles only two were shown to have an effect on creatine—creatinine excretion. The thyreotropic principle increased the excretion of creatine in the urine, and a preparation from the anterior pituitary which contains the follicle stimulating principle, increased the creatinine output.

Nitzescu and Gontzea (3) have recently published the results of their investigation on urinary creatine-creatinine excretion following the injection of certain preparations. The material they used was Prolan and Antuitrin S. (urinary gonadotropic principles), and Antuitrin G. (growth extract), and the subjects investigated consistently excreted creatine in the urine, apart from injections, which these authors recognised as a normal creatinuria. In this their preliminary communication, they class as normal the creatinuria occurring in subjects suffering from achondroplasic dwarfism, infantilism, gonadal insufficiency, and dwarfism. They claim that in such subjects prolan and antuitrin S. inhibit the creatinuria and exert a slight depressant effect on the excretion of creatinine; and that antuitrin G. increases the creatinuria and antagonises the effect of the urinary gonadotropic extract. Antuitrin G. injections into a normal adult were without effect.

The fact that our results differ from those of Nitzescu and Gontzea may be attributed to the fact that they were investigating abnormal subjects in which the endocrine system was undoubtedly involved, while we have confined our experiments to normal individuals.

The relation to acromegaly. Creatinine excretion in all cases of acromegaly as yet examined has been abnormally increased in the urine, and the responsible agent for raising this creatinine output appears, on experimental grounds, to be a gonadotropic principle secreted from the anterior lobe of the pituitary gland. Growth extract of the pituitary and anterior pituitary-like (Gonadotropic) substances produced no change in

urinary creatine or creatinine when injected into normal subjects. Although the factor promoting increased creatinine excretion appears to be inseparable from the gonadotropic principle of the pituitary in its preparation, future research with purer extracts will more certainly determine if this particular principle is itself the responsible agent. In acromegaly there are marked variations in the excretion of creatine in the urine. There may be a gross creatinuria persisting throughout the period of observation, or there may be a relatively low creatine output. Several cases of acromegaly appeared to have a diminished thyroid activity, for on examination the basal metabolic rate was depressed and creatine absent from the urine. The thyroid activity may be dependent on the secretion of the thyreotropic principle from the anterior lobe of the pituitary, as it has been shown in normals that injection of extracts containing thyreotropic principle produces obvious hyperthyroidism and increased creatine excretion. In all the cases we have investigated the high creatinine excretion has been a constant and characteristic finding. Excess of creatine is much more variable.

Gonadotropic extract injection in acromegaly. The pituitary extract was injected in doses of 5 ml. on five occasions, using two cases of acromegaly, with the typically raised creatinine excretion. No increase in creatinine excretion was produced, which was unlike the results obtained in normal subjects. These results, however, conform with those obtained by Schrire and Zwarenstein (6, 7) in rabbits which had been gonadectomised for a period of several months before injection with the gonadotropic principle. In such animals urinary creatinine is at a high level, and it has been suggested on experimental grounds that the post-castration hypertrophy of the anterior lobe of the pituitary is responsible for the altered creatinine output. In acromegaly, as in the long standing castrated rabbit, there appears to be a factor which eventually limits the quantities of creatinine in the urine. Because the provision of extra pituitary principle fails to increase creatinine excretion in acromegaly or in long standing castrated rabbits it would seem that the limiting factor cannot be connected with the pituitary secretions, but probably with the creatine stored in the muscles.

SUMMARY.

1. The effects of gonadotropic, thyreotropic, and growth extracts of the pituitary on the excretion of creatine and creatinine in the urine have been investigated in normal subjects.

2. Thyreotropic extract increases the elimination of urinary creatine by stimulating the thyroid gland to increased activity. Creatinine excretion is unaffected.

3. Gonadotropic pituitary extract increases creatinine excretion without affecting creatine in the urine. Urinary and serum gonadotropic extracts have no effect either on creatine or creatinine excretion. The factor increasing

creatinine elimination appears to be inseparable from the pituitary gonadotropic extract. Growth extract fails to alter the output of creatine or creatinine.

4. The excretion of creatine and creatinine in acromegaly is discussed.

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EXPERIMENTS RELATING TO THE ITCH SENSATION, IT'S PERIPHERAL MECHANISM, AND CENTRAL PATHWAYS.*

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WHILE scratching a recently inflicted gnat bite, I observed that while the itching appeared to originate at the point bitten, the surrounding skin also gave an abnormal itchy sensation when rubbed with the finger. A perfectly definite boundary could. be mapped between the normal and itchy skin. At this time the experiments of Lewis (5) on hyperalgesia were fresh in my memory, and it appeared to me that the distribution of this itchy skin resembled that of his hyperalgesia, and might be investigated along similar lines. The results of this investigation are recorded in Part 1 of this paper.

The itching sensation arising at the centre, has to be clearly distinguished from the surrounding itchy skin. I have therefore preferred to call it spontaneous itching ; because it continues to be felt without the intervention of a touch stimulus, which is necessary in the case of itchy skin. Spontaneous itching has been studied by Lewis, Grant, and Marvin (7) and is here considered only in relation to itchy skin. The central pathways through which both aspects of the itch sensation are conducted will be considered in Part 2.

Method.

The gnat bite is obviously inconvenient for the experimental study of itching. Itching powder has been tried, but it does not give a very pure form of itching, and it cannot be easily applied over a small area of skin. Histamine was finally chosen, since it has neither of these disadvantages. It has been punctured into the skin in strengths varying from 1 in 15 to 1 in 30,000 of the base dissolved in normal saline, according to the intensity and the duration of itching required. The stronger solutions have been buffered to p^H 7 to avoid any sensation arising from the introduction of an acid solution. Itchy skin, detected by the light friction of a finger, is mapped

out on the skin in ink, and traced on to cellophane for permanent record. All the experiments reported have been done on at least three subjects. An arm has been used in most experiments, the second being kept as a control.

Part 1. *The mechanism of itchy skin.*

After the puncture of histamine, there is a latent period of 20 to 30 sec. before the spontaneous itching begins. Itchy skin may usually be detected as a small area in the immediate vicinity of the puncture at the end of the first minute (Fig. 1). During the following 10 min. it continues to extend, forming an eliptical area. The long axis of the elipse always lies parallel to the cutaneous nerve trunks of the particular area chosen. The
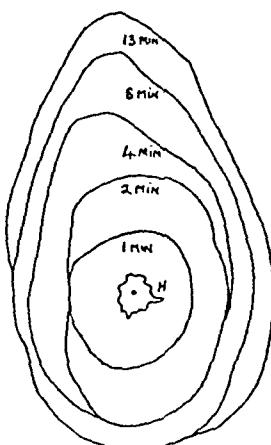

Fig. 1. (× ½). R.G.B., 19/5/37. Development of itchy skin from a histamine puncture. The point at which histamine was punctured, and the wheal which developed are shown at H. The areas of itchy skin, with the times at which they appeared, are incidated by the black lines.

0 min.	Histamine (1 in 15).
30 sec.	Spontaneous itching begins.
1 min.	Itchy skin first detected.
2 min.	Spontaneous itching intense.
7 min.	Spontaneous itching just perceptible. Itchy skin still extending.
17 min.	The whole area of itchy skin disappearing.

spontaneous itching rises to a maximum, declines, and disappears by the 10th minute. Thus itchy skin reaches its full extension after spontaneous itching has vanished ; it takes a further 10 to 20 min. to disappear.

Itchy skin could be explained by supposing it to result from diffusion of histamine from the puncture into the surrounding skin. But the fact that the areas are eliptical, and lie parallel to cutaneous nerves, suggests some kind of nervous control. This is proved by puncturing histamine into a small anæsthetic area (Fig. 2), when the appearance of itchy skin is delayed until the anæsthesia recovers. There is no delay if histamine is punctured

into a control injection of saline. The local novocain has blocked the nervous pathway involved in the production of itchy skin.

This pathway might be regarded as a local one in the skin, or as involving the spinal cord. The latter is excluded by the behaviour of itchy skin in relation to cutaneous novocain barriers (Fig. 3). Histamine is punctured into normal skin on the proximal side of the barrier. Itchy skin develops at a normal rate on this side, but it's appearance is delayed on the distal side until the barrier recovers. The close relationship existing between the
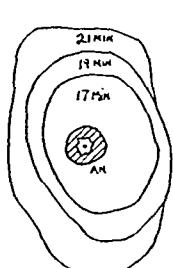

Fig. 2   Fig. 3.
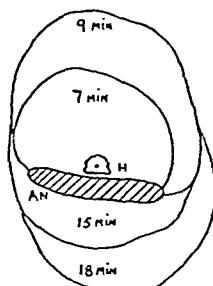
Fig. 2. (× ½). K.F., 16/6/37. Histamine punctured into anæsthetised skin.
    0 min.   Novocain (0·1 c.c. of 2%) injected into the skin giving area of anæsthesia (AN).
    1 min.   Histamine (1 in 15) punctured into middle of anæsthetic area.
  16 min.   No spontaneous itching and no itchy skin detected. Anæsthesia practically gone.
  17 min.   Spontaneous itching begins and first area of itchy skin mapped. No anæsthesia can be detected.

Fig. 3. (× ½). K.F., 16/6/37. The effect of a novocain barrier on the spread of itchy skin.
    0 min.   An oblong area of skin (AN) injected intradermally with novocain (0·25 c.c. 1%).
    5 min.   Histamine (1 in 15), punctured into the normal skin at H.
  10 min.   No recovery of sensation in the barrier can be detected. Itchy skin has not extended beyond it.
  15 min.   The barrier has now fully recovered sensation, and itchy skin has crossed.

position of a novocain barrier, and the development of itchy skin, may be further illustrated by puncturing histamine excentrically on an area of novocain anæsthesia (Fig. 4).

This experiment provides strong evidence against the participation of a central nervous mechanism, for the result would require cord reflexes of impossible complexity. Lastly, using Lewis's (5) argument, the failure of itchy skin to spread round the ends of the barrier may be taken as evidence in favour of a plexiform arrangement of the local nerves, rather than of a freely anastomosing network.

Other plexiform systems in the skin. This typical behaviour in relation to novocain barriers has now been shown to exist for goose-skin (Lewis and

Marvin (9), faradic sweating (Bickford (1)), and hyperalgesia (Lewis (5)). The local nerves in the first two cases are known to belong to the sympathetic system, whereas in itchy skin, like hyperalgesia, the sympathetic may be excluded.

The relevant experiment has been done on two patients with unilateral cervical sympathectomy (ganglionectomy). When histamine is punctured into the sympathectomised arm, the itchy skin produced is comparable in extent and duration to the control.
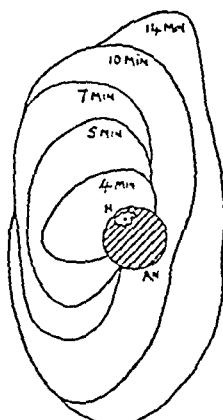

Fig. 4. (× ½). R.G.B., 11/5/36. The effect of puncturing histamine excentrically into anæsthetised skin.

  0 min.   Novocain (0·2 c.c. of 2%) injected intradermally and resulting area of anæsthesia (AN) mapped.

  2 min.   Histamine (1 in 15) punctured into margin of anæsthetic area at H.

  4 min.   The anæsthesia has receded slightly at its margins exposing the histamine puncture on one side. Adjacent to this the first area of itchy skin has developed.

  7 min.   The histamine puncture is now well outside the margin of anæsthesia. Itchy skin has developed only on the exposed side.

10 min.   The injected area has recovered sensation, and itchy skin is now extending beyond it.

Occurrence of itchy skin. Lewis, Grant and Marvin (7) showed that spontaneous itching is a feature of practically all forms of skin damage, provided that the damage is relatively slight in amount. Since itchy skin was unrecognised at this time these instances have now to be reinvestigated. This has been done for burns, freezes, mechanical pinching, galvanism, and ultra-violet burn. Itchy skin, similar to that produced by histamine has been found to follow each of these types of skin damage.

The detection of itchy skin in clinical cases of itching present some difficulties. The patient is first familiarised with the sensation of itchy skin produced by histamine puncture. The majority of patients soon learn to map the areas accurately. This sensation is now compared with that surrounding the clinical lesion. In all cases, patients have been quite certain that the itchy skin produced by histamine is identical with that surrounding

their own lesions. Scabies, flea bite, serum rash, carbolic dermatatis and herpes zoster commencing with itching, have been investigated in this way.

The peripheral process in itchy skin. It has been shown that the production of itchy skin depends on a local plexus of nerves. The sensation is conveyed to the central nervous system by sensory nerves. It would be consistent with contemporary ideas to postulate the liberation of a chemical intermediary between the two sets of nerve endings. This peripheral process must be highly unstable, since itchy skin shows no tendency to persist when the plexus nerves are blocked. Thus novocaine, injected locally at any interval following histamine puncture, leads to the rapid abolition of itchy skin. In this respect itchy skin differs from local hyperalgesia. The instability of peripheral process probably accounts for the fact that the existence of itchy skin cannot be prolonged by arresting the bloodflow to the part.
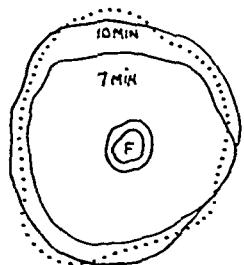

Fig. 5. (× ⅔). R.G.B., 27/5/37. Itching produced by a freeze.

   0 min. CO_2 snow (7 mm. diam.) applied to skin at F. for 20 secs..
   7 min.   Spontaneous itching begins and itchy skin is first detected ; the delay
            is due to cold acting as a local anæsthetic.
  10 min.   Spontaneous itching of moderate intensity. Itchy skin extending.
  12 min.   Spontaneous itching has become faint. A little soreness within the
            area of itchy skin.
  16 min.   Slight burning pain has replaced the spontaneous itching. The
            surrounding skin now definitely hyperalgesic. It's boundary is
            shown by the dotted line.

The relation of itchy skin to hyperalgesia and the flare. In certain subjects the itchy state of skin following histamine puncture changes to a state of hyperalgesia. The hyperalgesia is first detected 10 to 15 minutes after puncture, and spreads to occupy fully the area previously displaying an itchy condition. It persists for many hours. When itching is produced by skin damage, all subjects seem to show this transition from itchy skin to hyperalgesia. The spontaneous sensation often undergoes a corresponding change from itching to burning pain (Fig. 5).

The final outline of the vascular flare coincides approximately with that of itchy skin. Yet itchy skin is not the result of the flare, for it can be produced in an arm after arrest of its circulation.

The close relation between the outline of itchy skin, flare and hyperalgesia would be explained if they were mediated through a single system of plexus nerves. But Lewis (6) has already shown that the nerves mediating flare and hyperalgesia differ in regard to their sensitivity to blocking by asphyxia. The nerves mediating itchy skin show a greater difference in this respect, for itchy skin crossed an area rendered anæmic by the pressure of a rubber band applied for many hours; while in Lewis's experiments spread of hyperalgesia was prevented by 20 to 30 minutes, and spread of flare by a little over 30 minutes, pressure.

Part 2. *The central pathways involved in itching.*

In the preceding account, the sensations of both spontaneous itch and of itchy skin have been assumed to be conveyed through a single system of sensory nerves. However, the fact that in the one case the sensation arises spontaneously, whereas in the other it only exists in association with a touch stimulus, might lead us to expect some kind of differentiation in the nerves. Experiments now to be recorded, show that the nerves carrying the two sensations are different in their properties.

The effect of asphyxia on itch nerves. Lewis, Pickering and Rothschild (9) have shown that when the circulation to an arm is arrested by means of a forearm cuff, loss of sensation begins distally, and spreads in a proximal direction. Thus at any particular moment the arm may be divided into zones of sensory dissociation. These changes are a result of a graded sensitivity to asphyxia exhibited by the nerves lying beneath the cuff. Since the itch nerves also share in the asphyxiation the preparation may be used to detect any variation in sensitivity which they exhibit.

The circulation to the whole limb is arrested until anæsthesia spreads to the dorsum of the hand; sensation is normal in the vicinity of the cuff. This usually takes about 25 minutes. Histamine is punctured into the anæsthetic area, the skin which retains normal sensation, and a control area on the other arm. Spontaneous itching occurs equally at the three punctures. Itchy skin fails to appear in the anæsthetic area but occurs in the skin with unimpaired sensation. This observation shows that the failure of itchy skin to appear is due to asphyxial blocking of the sensory nerves at the cuff, and not the local plexus nerves in the skin; for the latter would be equally affected throughout the arm. By placing histamine punctures in position intermediate between the normal and anæsthetic skin, it can be shown that itchy skin fails to appear when sensory impairment is at a less advanced stage than complete loss of touch. In fact it fails at a much earlier stage when the sensation of tickle* is lost. The latter is tested by means of light strokes with a wisp of cotton wool, and can be clearly distinguished from the more advanced stage of hypoæsthesia, in which only

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* Blake Pritchard (2) has called this sensation, " superficial tickle."

the heavier touches of a finger are appreciated. Thus, the nerves conveying the itchy skin and tickle sensations share the property of being blocked at an early stage by asphyxia.

Spontaneous itch has already been shown to remain unimpaired in skin which has lost the sensation of touch. It remains so until, at a very late stage in asphyxia, the pain sensation is practically lost. This may be shown by arresting the circulation to a single finger, a method used by Lewis and Pochin (10). In this case the sensation of itchy skin with that of tickle is lost in about 45 minutes. By the end of the first hour the finger has become anæsthetic. The spontaneous itching from histamine remains practically undiminished until about $1\frac{3}{4}$ hours. Spontaneous itching subsequently becomes gradually reduced in intensity, until at 2 hours, just before the finger has become analgesic, it fails altogether.

Thus the fibres conveying the sensation of itchy skin and of spontaneous itching, present a contrast in their sensitivity to blocking by asphyxia. The former are grouped with the fibres conveying tickle in being the most sensitive, and the latter with those of pain in being among the most resistant to the effects of asphyxia.

Effects of cooling nerve. These results received further support from experiments on nerve cooling. It is intended to record these fully in a subsequent publication. A forearm nerve is cooled by perfusing a freezing mixture at -5°C. through a lead pipe lying on the skin immediately over a small length of it. Sensation becomes impaired in an area of skin well distal to the point cooled. As cooling proceeds sensation is lost in the same order as that produced by asphyxia ; namely, tickle first, followed by touch and later by pain. The loss of sensation is not due to asphyxia since it recovers immediately on rewarming the nerve when the circulation to the arm is arrested.

When histamine is punctured into the area and cooling of the nerve is begun the itchy skin disappears early at the same time as superficial tickle. As in the case of asphyxia, spontaneous itching remains till a later stage. It is last felt just before the onset of complete analgesia.

Histamine itching in clinical cases of sensory dissociation. Many workers (Thöle (11), Erhenwald (3), Blake Pritchard (2)) have investigated the occurrence of itching in various clinical cases of sensory dissociation. It is difficult to incorporate their results because the two types of itch sensation were not recognised at the time. Thus itching is often referred to as " qualitatively altered," when it is clear that the state was one of itchy skin in the absence of spontaneous itching. Many statements are made concerning the relation of itching to the touch sensation. Blake Pritchard distinguished clearly between tickle and touch, this distinction is a very important one.

The occurrence of itching in a small series of cases of sensory dissociation has been reinvestigated. The patient is first familiarised with the sensations of spontaneous itching and itchy skin by puncturing histamine into a normal

part of the skin.   In the area of sensory dissociation particular note is taken of the presence of tickle, touch, and pin prick pain sensations.   Histamine is punctured into the middle of such an area and the occurrence of itchy skin or spontaneous itching is noted and compared with a control.   The results are shown in Table I.   They may be summarised.

Itchy skin cannot be detected in skin which has lost tickle sensation, although touch may be retained (syringomyelia and cordotomy cases). Where tickle is impaired, itchy skin often appears as a reduced area, or it may fail altogether.   The close relation between tickle and itchy skin noted during the asphyxia and cooling experiments, is corroborated.

Spontaneous itching is not felt where pain to pin prick is dulled.   It was conspicuously increased in one case of nerve damage where the response to pin prick was exaggerated.

TABLE I.

Name.	Diagnosis.	Position of histamine puncture.	Tickle.	Touch.	Pain.	Spontaneous itching.	Itchy skin.
H.M.	Paraplegia from cord tumour	(1) Legs	absent	present	dull	absent	absent
		(2) Below umbilicus	reduced	present	dull	absent	small area
H.B.	Brown Séquard syndrome	Abdomen	slight	present	absent	absent	small area
W.C.	Injury radial nerve	(1) Knuckle	absent	absent	increased	increased	absent
		(2) Dorsum of hand	slight	slight	dull	absent	absent
A.C.	Syringomyelia	(1) Shoulder	absent	present	dull	absent	absent
		(2) Face	absent	slight	absent	absent	absent
G.M.	Syringomyelia	Infraclavicular	absent	present	absent	absent	absent
E.M.	Cordotomy	Legs	absent	present	absent	absent	absent

The case of cordotomy is of particular interest.   Preliminary testing showed that there was complete loss of pin prick and tickle over the affected area.   The touch sensation was quite intact.   Histamine puncture produced neither spontaneous itching, nor itchy skin.   Since in this case only the antero-lateral tracts were divided, these results must mean that the four sensations, tickle, itchy skin, spontaneous itching, and pain are carried by it. This probably accounts for the fact that these four sensations tend to disappear together in cases of spinal tumour and syringomyelia although touch in the same area may remain little affected.

Antipruritic state. Lewis, Grant, and Marvin (7), were first to report the occurrence of a peculiar state of the skin, following a faradic stimulus, in which puncture of histamine failed to produce itching. This state, which may be conveniently termed "antipruritic," is not peculiar to faradism but may be produced by a large number of painful stimuli. It seemed to be of sufficient interest to merit further experimental study.

The state may be most conveniently induced in the skin, by applying for 3 min. the end of a metal rod (1 cm. diam.) maintained at 40°C. by means of a water jacket. Histamine punctured within 10 cm. of the heated area, produces no spontaneous itching or itchy skin. There may be a reduction in their intensity and duration over a much more extensive area. Recovery from this state takes about 6 hours.

A similar antipruritic state may be produced by a painful galvanic stimulus, or mechanically, by a strong blow on the skin with a ruler. The state has been found to be antipruritic towards itching produced by various methods (gnat bite, burn, freeze, and galvanism).

The antipruritic state might arise locally from the direct stimulation of nerve endings, or through the intermediary of a chemical substance. In the latter case, the application of the stimulus on a small area of anaesthetic skin should only delay the onset of the state until the anaesthetic had recovered. Repeated experiment has shown that the antipruritic state is not produced under these conditions. It is, therefore, concluded that the stimulus producing antipruritic state does so through a direct action on the nerve endings.

The antipruritic state might be produced through local nervous pathways or those involving the spinal cord. The following experiment shows that the latter hypothesis is correct. Two adjacent forearm nerves are injected with novocain and the resulting area of anaesthesia is mapped. A strong faradic stimulus is laid down in the middle of this area, and at a corresponding point on the other arm. When the anaesthesia recovers (about ¼ hour) histamine is punctured on either arm within an inch of the points faradised. Normal itching occurs on the arm which was anaesthetised, but the control produces no itching since it has become antipruritic. An adequate stimulus has in this instance failed to produce the antipruritic state although the local nervous pathways remained open. It must, therefore, be produced through a spinal cord mechanism.

The varied stimuli that will produce an antipruritic state have in common the property of being painful. This suggests that these stimuli may act through the pain pathway. The antipruritic state can in fact be induced in skin which only has pain sensation intact. The experiment is done on a finger which has had the circulation arrested by means of a rubber band for 1¾ hours. At this time practically all sensation except pain has been abolished, yet a normal antipruritic area is produced on faradising the finger.

Summary.

(1) When histamine is punctured into the skin itching begins at the point of the puncture. The skin widely surrounding the puncture also undergoes a change, so that when gently rubbed it gives an abnormal itchy sensation. The former sensation is called spontaneous itching, and the latter itchy skin.

(2) Itchy skin arises through a local nervous (axonic) pathway. This pathway is separate from that of hyperalgesia and the vascular flare. The nerves are arranged in a plexiform manner. They do not belong to the sympathetic system.

(3) Itchy skin has been found around all itching lesions examined, whether occurring naturally or produced deliberately. It is, therefore, considered to be an essential part of the itch sensation.

(4) Itchy skin may be abolished by a degree of asphyxia which leaves spontaneous itching unaffected ; a similar dissociation may be produced by cooling a nerve trunk. This indicates that the nerves carrying the two sensations are separate. The tickle sensation shows a close association with itchy skin in these experiments.

(5) In clinical cases of sensory dissociation, itchy skin is not detected in areas where tickle sensation is lost, even though touch may be retained. Spontaneous itching is not felt where pain to pin prick is defective. Tickle pain, spontaneous itching, and itchy skin are all conveyed in the antero-lateral tracts, since they all disappear when this is divided.

(6) A state of skin in which itching is inhibited has been described. The inhibition is central in origin, and is probably produced through the pain nerves.

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SOME OBSERVATIONS ON CLUBBED FINGERS.

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CLUBBED fingers are found associated with such diverse conditions as pleuro-pulmonary diseases of all kinds, congenital heart disease, subacute bacterial endocarditis (4), post-thyroidectomy myxœdema (1, 23), cirrhosis of the liver, especially of the biliary type (2), and various chronic diarrhœas, such as sprue (10), amœbic dysentery (12) and non-specific ulcerative colitis (23). They may occur in the absence of these diseases as an hereditary abnormality, transmitted as a Mendelian dominant (9, 24). They may also be found in one extremity only, usually accompanying an aneurysmal dilatation of the artery to that extremity (7, 14, 18).

The immediate cause of the clubbing may be (1) a nervous mechanism, (2) a generalised chemical disturbance, manifesting itself in the finger tips or (3) a circulatory disturbance. Assuming a single mechanism for all cases, it is difficult to explain unilateral clubbing on a chemical basis and because of the absence of demonstrable nerve lesions, difficult to explain bilateral clubbing on a nervous basis. This paper describes an investigation into the remaining possibility, namely, a disturbance in circulation.

To investigate the possibility that the bloodflow through all or a part of the fingers is altered in clubbing, skin temperatures were measured thermoelectrically with the unit described by Lewis (13) or the portable modification of Grant (5). Skin temperatures measured from the exposed hands of subjects at rest in bed showed very wide variations, as did the air temperatures to which they were exposed ; and it was impossible to establish any difference between the finger temperatures of subjects with and without clubbing in this way. Skin temperatures measured from the tips and bases of the fingers were also very similar in normal and clubbed fingers, when sympathetic vasoconstrictor tone had been removed from the hand by immersing the other arm in water at 43 to 45°C. for about 30 minutes, a time sufficient to allow the finger temperatures to reach a plateau. Skin temperature is not, however, especially at this plateau level, a very delicate index of bloodflow.

A more delicate method for estimating bloodflow under these conditions is that described by Stewart (22). Accordingly, the heat elimination from the hand, from which vasoconstrictor tone had been removed by warming the body, was measured in 8 cases of clubbing, using the procedure described by Pickering (20). The heat elimination measured under these conditions is referred to throughout this paper as the maximum heat elimination. Table I compares the maximum heat elimination from the hand of the

TABLE I.

Maximum heat elimination of the hand.

Control subjects (Pickering).				Clubbed finger cases.*			
Subject.	Age and sex.	Calories per100c.c. per min.	Diagnosis.	Subject.	Age and sex.	Calories per100c.c. per min.	Diagnosis.
H.S.	17 M.	106	Normal.	M.M.	16 M.	77	Bronchiectasis.
M.W.	22 F.	130	Synovitis.	J.C.	17 M.	57	Bronchiectasis.
E.B.	24 M.	108	Normal.	M.C.	22 M.	122	Bronchiectasis.
G.B.	26 F.	130	Pyelitis.	E.R.	30 F.	133	Bronchiectasis.
G.P.	31 M.	100	Normal.	C.T.	38 M.	45	Pul. tuberculosis.
J.S.	31 M.	87	Gastritis.	H.P.	43 M.	87	Empyema.
P.R.	34 M.	110	Normal.	E.T.	45 M.	67	Empyema and bronchitis.
F.B.	40 F.	129	Normal.	W.L.	50 M.	68	Empyema.
F.M.	44 M.	71	Gastric ulcer.				
W.H.	47 M.	90	Gastric ulcer.				
A.H.	51 M.	122	Gastric ulcer.				
H.H.	51 F.	78	Hernia.				
A.W.	52 F.	128	Biliary cirrhosis.				
R.A.	54 M.	118	Cholecystitis.				
J.H.	56 M.	63	Gastric ulcer.				
R.D.	58 M.	118	Gastric ulcer.				
D.B.	58 F.	79	Diabetes.				
A.M.	62 M.	42	Carcinoma of stomach.				
E.B.	62 F.	68	Gastritis.				
A.D.	63 F.	89	Diabetes.				
F.D.	69 F.	83	Retinal thrombosis.				

* Studied through the courtesy of Prof. F. G. Chandler.

patients with clubbed fingers and the maximum heat elimination obtained in the same way by Pickering (20) in a series of normal subjects. Although the values for maximum heat elimination per 100 c.c. of hand volume in the subjects with clubbed fingers fall within the normal range, this range is very

wide. It is clear, therefore, that any abnormality in bloodflow confined to the finger tips may not be demonstrable in measurements of heat elimination from the whole hand.

To measure the heat elimination from the finger tip the following calorimeter was devised (Fig. 1).
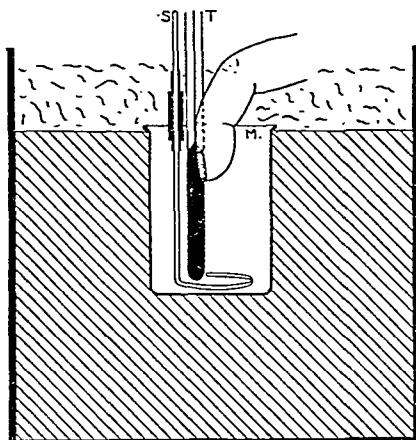

Fig. 1. (× 1/3). A rimmed polished aluminium cup, 4·5 cm. across and 6·5 cm. deep, is covered with a piece of thin rubber (M), moderately stretched over the rim and securely tied. A hole, approximately 1 cm. across, is burned through the centre and two smaller holes, about 3 mm. each across and 1 cm. apart, are burned near the edge of this rubber diaphragm. The burned edges are wiped clean with ether. To stir, a glass rod (S) is used, to the bottom of which is attached at a right angle a piece of thick copper wire bent to the arc of a circle of slightly smaller radius than that of the cup. A gap between the free end of the wire and the rod allows space for the bulb of the thermometer (T). This stirrer is inserted through one of the lateral holes in the rubber dam and a piece of close fitting glass tubing is slipped over the rod and is fixed in the hole of the rubber diaphragm by a piece of rubber tubing. The rod is moistened with a little glycerine, which as it mixes with the water makes a good lubricant.

The cup is surrounded with broken cork in a box (16 × 16 × 16 cm.). A large thermometer graduated to hundredths of a degree is suspended from a ring stand into the cup, passing through the other lateral hole in the rubber diaphragm to the bottom of the cup, the bulb of the thermometer being thus completely below the diaphragm. The cup is now filled to the top with water at a temperature such that the final temperature of the calorimeter and its contents is about 30·0°C.. 5 c.c. of water is then removed.

The patient is sitting up in bed and before and during the period of calorimetry his left arm is immersed in stirred water at 43 to 45°C.. When the patient begins to sweat (15 to 25 minutes), a finger, usually the fourth of the right hand, is inserted into the central hole in the rubber diaphragm down to the first volar crease. The position of the forearm, wrist and hand, as well as that of the box, is adjusted so that the arm rests comfortably on the edge of the bed and the finger hangs perpendicularly into the cup. Cotton wool is packed loosely around the hand and over the cup and broken cork.

Stirring is now begun at the rate of about 200 strokes per minute and after a few minutes the rise in temperature per minute is recorded. The

first reading is usually taken at about 30·40°C. and the last at about 31·60°C..
The rise is steady, tending to be slightly less at the upper than at the lower
level of temperature.   The measurements are usually made over a period of
from 6 to 10 minutes, depend on the rate of temperature rise.   At the end
of this time the finger is removed, the level of immersion marked with ink
and a small cork inserted into the central hole in the diaphragm.   Stirring
is again begun and the fall in temperature per minute recorded over a period
of 5 minutes.   If the room temperature is above 20°C., the fall is usually
0·06° to 0·08°C. per minute, although at temperatures below 20° it may be
as great as 0·10°C. per minute.   The surface area of the finger is measured
by covering the immersed part with strips of adhesive tape, the areas of
which are subsequently measured.   The volume is determined by noting
how much water the immersed finger tip displaces.   The volume of the water
in the calorimeter is also measured (98 to 104 c.c.).

The maximum heat elimination of the finger tip is calculated from the
following formula : $M H E = (W + W E) \times (H + h)$, in which W is the
volume of water in the cup, W E is the hydrothermic equivalent of the cup,
the wire and the thermometer bulb (the glass rod is neglected), H, the average

TABLE II.

Maximum heat elimination of the finger tip.

Subject.	Date.	MHE. per c.c. per min.	MHE. per sq. cm. per min.	Diagnosis.
H.M.	16.6.38	4·67 cal.	1·65 cal.	Normal.
	19.7.38	4·81	1·63	
A.H.	19.7.38	3·81	1·47	Normal.
	20.7.38	4·07	1·32	
P.W.	24.5.38	3·80	1·51	Normal.
	10.8.38	3·85	1·47	
A.L.	20.5.38	3·61	1·49	Normal.
	20.6.38	3·78	1·39	

rise in temperature per minute and h, the average fall in temperature per
minute after the finger has been removed.   M H E is divided by the surface
area for calories per sq. cm. per minute and by the volume for calories per c.c.
per minute.   Table II shows the M H E of four normal subjects, each
estimated on two occasions.

Table III shows the M H E of clubbed and normal fingers.   The normal
range is much narrower for the finger than for the hand.   The measurements

per unit surface area, moreover, are less variable than those per unit volume. They are also more relevant because only the surface and not the deeper tissues are concerned in heat elimination. The maximum heat eliminations of clubbed fingers secondary to pleuro-pulmonary and congenital heart disease are in most instances definitely above the upper limits of normal, although there are a few which fall within the upper normal range. We can conclude from this that the bloodflow per unit surface under these conditions is greater in the clubbed than in the normal finger tip. No relationship can be established between the degree of increase in flow and the degree of clubbing. In fact, some of the highest readings were obtained in fingers with only slight clubbing.

TABLE III.

Maximum heat elimination of the finger tip.

Subject.	Age and sex.	Mouth temp.	Pulse rate.	MHE. per c.c. per min.	MHE. per sq.cm. per min.	Clubbing.	Diagnosis.
P.W.	18 M.	99·4°F	84	3·80 cal.	1·51 cal.	0	Normal.
D.S.	18 M.	99·4	88	3·96	1·40	0	Normal.
B.B.	19 F.	99·6	88	6·14	1·83	0	Normal.
A.L.	19 M.	98·8	76	3·78	1·39	0	Normal.
S.R.	23 M.	99·2	92	4·78	1·76	0	Normal.
A.R.	23 M.	99·2	92	4·55	1·74	0	Normal.
M.W.	24 M.	99·4	76	6·19	1·80	0	Normal.
W.P.	25 M.	99·2	72	3·62	1·43	0	Normal.
A.M.	30 M.	99·4	88	5·13	1·87	0	Normal.
M.M.	30 M.	99·3	98	4·67	1·65	0	Normal.
A.H.	30 M.	99·2	80	4·07	1·32	0	Normal.
C.B.	20 M.	99·0	104	6·11	1·93	+++	Congenital heart disease.
T.B.	22 M.	99·6	108	6·15	2·09	+++	Bronchiectasis.
D.N.	22 F.	99·5	120	6·05	2·23	++	Lung abscess.
D.C.	26 F.	99·6	108	6·35	2·43	+	Bronchiectasis.
N.M.	30 F.	99·7	100	5·77	2·08	+	Bronchiectasis.
L.B.	32 M.	99·6	104	5·09	1·81	++	Empyema.
R.G.	36 M.	98·8	88	5·17	2·09	++++	Bronchiectasis.
F.T.	36 M.	99·6	132	4·37	1·60	+++	Empyema Hypertrophic osteoarthropathy.
S.P.	38 M.	100·1	120	5·82	2·08	++	Pulmonary tuberculosis.
J.B.	46 M.	99·0	108	4·91	1·98	++	Bronchiectasis.
A.N.	50 M.	100·2	116	5·09	1·61	++	Lung abscess.

Since the bloodflow in the finger tip is directly proportional to the digital arterial pressure and inversely proportional to the resistance in the digital vessels, it seemed desirable to compare the digital arterial pressures of normal with those of clubbed fingers. The digital systolic arterial pressure may be measured by the method of Gaertner (3). A metal cuff lined with a rubber membrane is fitted over the middle phalanx of the finger (usually the 4th) after it has been blanched by rolling a tight rubber band over it to the base.

The cuff is inflated to a pressure above systolic and the rubber band is severed. As the pressure is slowly lowered the point is noted when the tip of the finger suddenly flushes. The diastolic pressure may also be measured with Gaertner's capsule. When the blood pressure is taken in the arm, throbbing usually starts somewhere above systolic pressure but always stops abruptly at the diastolic pressure as measured by the auscultatory method. When, in the warm finger, the pressure in the Gaertner capsule is gradually reduced from above systolic pressure, throbbing becomes distinct and then abruptly disappears as the pressure falls. The point at which throbbing disappears is taken as the diastolic pressure in the digital artery.

Table IV summarises pressures recorded from the upper arm by the auscultatory method and from the finger by the methods described. All these pressures represent readings obtained from one upper extremity after the other had been immersed in hot water long enough to produce generalised cutaneous vasodilatation. Both systolic and diastolic pressure gradients from the arm to the finger were definitely less than normal in all cases of clubbing associated with lung or congenital heart disease. The difference

TABLE IV.

Arterial blood pressure after warming.

Subject.	Age and sex.	Brachial mm.Hg.	Digital mm.Hg.	Gradient (mm.Hg.)		Clubbing	Diagnosis.
				Systolic.	Diastolic.		
P.W.	18 M.	102/74	88/40	14	34	None	Normal.
D.S.	18 M.	125/84	96/52	29	32	,,	Normal.
B.B.	19 F.	135/94	110/76	25	18	,,	Normal.
A.L.	19 M.	114/82	106/52	8	30	,,	Normal.
S.R.	23 M.	132/86	116/60	16	26	,,	Normal.
A.R.	23 M.	112/82	84/70	28	12	,,	Normal.
M.W.	24 M.	128/80	102/60	26	20	,,	Normal.
W.P.	25 M.	118/85	102/64	16	21	,,	Normal.
R.W.	25 M.	116/84	85/40	31	44	,,	Normal.
G.S.	26 M.	150/76	110/66	40	10	,,	Graves' disease.
E.B.	28 F.	130/76	98/25	32	51	,,	Anæmia.
A.M.	30 M.	118/90	96/60	22	30	,,	Normal.
M.M.	30 M.	116/86	98/54	18	32	,,	Normal.
A.H.	30 M.	124/90	100/35	24	55	,,	Normal.
C.H.	32 M.	135/95	104/76	31	19	,,	Hæmophilia. No anæmia.
W.C.	32 M.	210/160	210/155	0	5	,,	Malignant hypertension. Acromegaly.
A.M.	32 M.	124/82	105/45	19	37	,,	Normal.
F.C.	34 F.	175/130	148/104	27	26	,,	Essential hypertension.
P.S.	36 F.	155/100	115/75	40	25	,,	Essential hypertension. Erythrocyanosis.
C.M.	42 M.	122/84	100/56	22	28	,,	Neurasthenia.
E.H.	54 F.	210/135	180/95	30	40	,,	Essential hypertension.
E.C.	55 F.	165/120	135/100	30	20	,,	Essential hypertension.

TABLE IV *(continued)*.

Subject.	Age and sex.	Brachial mm.Hg.	Digital mm.Hg.	Gradient (mm.Hg.)		Clubbing	Diagnosis.
				Systolic.	Diastolic.		
C.B.*	20 M.	102/88	92/66	10	22	+++	Congenital heart disease.
D.R.	21 F.	105/72	88/64	17	8	+	Pulmonary tuberculosis.
T.B.	22 M.	104/74	104/58	0	16	+++	Bronchiectasis.
D.N.	22 F.	125/74	116/66	9	8	++	Lung abscess.
A.V.	24 F.	128/86	110/84	18	2	++	Bronchiectasis? Raynauds' disease. Bilateral sympathectomy.
D.C.	26 F.	136/92	128/94	8	−2	+	Bronchiectasis.
M.D.	28 F.	155/110	138/90	17	20	++	Lung abscess.
J.M.	30 M.	130/85	116/86	14	−1	+	Lung abscess.
E.R.	30 F.	105/70	100/55	5	15	++	Bronchiectasis.
N.M.	30 F.	128/90	122/74	6	16	+	Bronchiectasis.
L.B.	32 M.	138/80	124/72	14	8	++	Empyema.
T.P.	35 M.	110/80	95/75	15	5	+++	Empyema.
F.T.	36 M.	124/68	95/65	29	3	+++	Empyema. Hypertrophic osteoarthropathy.
R.G.	36 M.	142/90	126/80	16	10	++++	Bronchiectasis.
S.P.	38 M.	112/80	108/74	4	6	++	Pulmonary tuberculosis.
P.A.	43 M.	200/120	160/75	40	45	++++	Essential hypertension. Hereditary clubbing.
W.B.	45 M.	130/84?	102/78?	28	6	++	Mitral stenosis. Auricular fibrillation. Congestive heart failure. Bronchiectasis.
J.B.	46 M.	136/90	124/76	12	14	++	Bronchiectasis.
A.N.	50 M.	126/98	102/82	24	16	++	Lung abscess.
W.L.	50 M.	126/78	110/85	16	−7	+++	Empyema.
J.A.	54 M.	140/90	110/35	30	55	++++	Hereditary clubbing.

* These cases were studied through the courtesy of Mr. J. E. H. Roberts, Mr. A. J. Morland and Mr. R. S. Pilcher.

between these gradients was due to abnormally high digital rather than to low brachial pressures in patients with clubbed fingers. The degree of elevation of digital arterial pressure was usually correlated with the degree of increase in maximum heat elimination. In two cases of hereditary clubbing,* the gradients were normal, despite the co-existence of hypertension in one of these cases. In 3 patients with essential hypertension, the gradient was normal, but in one patient with malignant hypertension complicating acromegaly, the gradient was much decreased. Similar findings in several cases of malignant hypertension with relation to the systolic gradient were reported by Oppenheimer and Prinzmetal (19). In all the other cases tested, such as anæmia and Graves' disease, the gradients were within normal limits.

* Studied through the courtesy of Dr. D. W. Seaton of Leeds.

Clubbed fingers occurring symmetrically in patients with lung or congenital heart disease thus seem to differ from the normal in two features. First, when sympathetic vasoconstrictor tone is removed from the fingers by raising body temperature, the rate of bloodflow through the finger tip is abnormally great. Second, under the same circumstances, the fall of pressure from brachial to digital arteries is abnormally small.

In addition to the studies made on cases of bilateral clubbing of the fingers, an opportunity was presented of studying 4 cases of unilateral clubbing of the fingers. Because of their special interest, these cases are reported in detail.

Case 1*. G.A., a man of 59 years, was admitted in June, 1937, suffering from an aneurysm presenting in the right subclavian triangle. He complained of pain in the right shoulder of 6 months' duration, the pain having spread to the arm and hand in the last 3 months. The pain was described as a severe and continuous ache. He had also experienced pins and needles and numbness of all the right fingers, but especially the 4th and 5th fingers. Tenderness of the fingers, swelling of the hand and increased curvature of the nails had been noticed for 3 months.

The aneurysm presented itself as a large, firm, tender swelling, showing expansile pulsation in the right subclavian triangle. The heart presented few signs of enlargement, but there was retrosternal dulness at the level of the second interspace. A roentgenogram showed the aortic knob to be normal; above this an abnormal shadow projected to the right and left of the sternum, corresponding to the dulness. A roentgenogram taken 14 months previously showed no such shadow. The pulse was regular and equal in the two arms, and the brachial blood pressures were 130/75 on the left and 125/80 on the right. The Wassermann reaction was positive.

The right arm was swollen. The veins of the right arm were more prominent than those of the left. Pitting was just distinct on the hand and arm as high as the elbow. A slight general swelling of the hand was noticed; swelling of the right fingers was obvious. On the right side the wrinkles of the skin of the fingers at the joints were much reduced, and the skin was tender. All five fingers of the right hand were clubbed. The increased curvature of the nails is well displayed in the photographs (Fig. 2 and 3). Measurements of the nails showed all 5 to be increased in length and breadth on the right side, the differences varying between 3 and 6 mm.. A roentgenogram of the hands showed rarefaction of all the right bones; there was no other change except measurable shortening of 2 terminal phalanges on the right side.

The man stated, and observation confirmed him, that the right hand was ordinarily warmer than the left. The hands were exposed symmetrically on the bed, after being thoroughly and equally warmed beneath the clothes; at the end of 35 minutes, the right hand and arm were still quite warm, but

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* I am indebted to Sir Thomas Lewis for these notes of his observations on this case.

the left hand and arm were obviously colder. This test was repeated with similar result on several subsequent days.

In this case the decreased cooling of the involved hand in response to low environmental temperature was probably a manifestation of involvement of the sympathetic nerves to the upper extremity. It is to be noted that there was also venous and perhaps lymphatic obstruction.

Case 2. W.B., a man of 50, was admitted to the hospital complaining of breathlessness on slight exertion and attacks of precordial pain radiating to the left shoulder and down the left arm of 8 months' duration. For a month he had also experienced a more steady aching pain in the right shoulder.

The patient presented the signs of free aortic regurgitation, namely, waterhammer pulse, enlarged heart, and basal diastolic murmer. There were râles at both lung bases. The Wassermann test was positive. A roentgenogram of the chest showed enlargement of the left ventricle and dilatation of the aorta. There was moderate clubbing of all the fingers of the right hand and some shortening of the distal phalanges. The left hand was normal in appearance, except for a partial amputation of the distal phalanx of the index finger due to an accident at work.

The temperatures of the dorsal surfaces of the distal phalanges of the third finger of each hand were measured thermo-electrically. No significant differences were found between the two sides either under ordinary conditions or after the body had been thoroughly warmed by placing both feet in water at 43 °C. for $\frac{1}{2}$ an hour. The bloodflow to the fingers was measured in both hands by the method described by Grant (6) at a bath temperature of 31 °C.. The volume flow was moderately less in the clubbed fingers of the right than in the fingers of the left hand. The maximum heat eliminations of the tips of the 4th fingers of both hands were measured by the method already described. On the right, the readings were 2·90 calories per c.c. per minute and 0·97 calories per sq. cm. per minute. On the left they were 3·89 calories per c.c. per minute and 1·16 calories per sq. cm. per minute. The pulsations of the radial and brachial arteries were definitely greater on the right than on the left side. Because of the aortic insufficiency, it was difficult to determine the brachial diastolic pressures of either arm exactly. Under ordinary conditions the brachial and digital pressures on each side were as follows :—

	Brachial.	*Digital* (4th)	*Systolic gradient.*
Right	190	165	25
	?	75	
Left	180	150	30
	?	70	

After warming the body the pressures were :—

Right	165	125	40
	?	65	
Left	150	125	25
	?	65	

It is apparent here that the maximum bloodflow was below the limits of normal in the fingers of both hands and lower in the clubbed than in the normal. There were, on the other hand, no essential differences in the digital arterial pressures under the same conditions.

Case 3.* M.M., a 60 year old housewife, complained of moderate aching pain in the left forearm and hand. The left arm and hand had been swollen, the veins of the left forearm prominent, and the finger nails of the left hand, curved as long as she could remember. In various regions of the forearm and hand she had developed " tumours," which grew larger and ached when the hand was dependent or cold. Sometimes a " tumour " became discoloured, more swollen, and acutely painful and tender for a few weeks. When she was a child, one of these swelling burst within the forearm, which became very swollen and irregularly blue. Eight years before admission a lipoma in the left scapular region, together with several bluish grape-like excrescenses in the left axilla were removed surgically. The scapular tumour recurred and was removed 4 years later.

The entire left forearm and hand was larger than the right. There was slight pitting on pressure in the wrist and hand. The veins of the arm and forearm especially on the dorsal surfaces were much dilated. Valves were demonstrable in the normal veins on the volar aspect of the forearm but not in the dilated ones on the dorsal aspect. There were several swellings ranging in size from a plum to a grape in the subcutaneous tissues of the left forearm and hand. Some of these were firm and fibrous whereas others were soft and fluctuating. Some of the softer ones had a bluish tint. When a cuff was applied to the arm at a pressure of 30 mm. of mercury, the swellings became engorged with blood. The index and middle fingers of the left hand were conspicuously clubbed (Fig. 4). The thumb and ring finger were only moderately clubbed, whereas the 5th finger did not appear to be clubbed at all. The nail beds were cyanotic and more deeply cyanotic in the more clubbed fingers. Under the nail of the index finger there was a splinter hæmorrhage which subsequently disappeared. The X-ray revealed calcifications in the soft tissues of the hand as well as an increase in width and length of the distal phalanges of the index and middle fingers. When the patient was seen 6 months later she complained of pain below the left elbow. This was apparently due to a thrombosis of a varicosity.

When the hands were equally exposed, no difference in temperature between the two was discernable from palpation or from thermo-electric measurements. The left hand, however, cooled somewhat more quickly than the right, due probably to its increased surface area. There were no significant differences in the heat eliminations of the two hands. The

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* I am indebted for this case to Miss A. L. Winner.

maximum heat eliminations of the right and left third finger tips were as follows :—

Right—1·35 calories per sq. cm. per minute.
3·95 calories per c.c. per minute.

Left 1·49 calories per sq. cm. per minute.
3·23 calories per c.c. per minute.

The volume of the left finger tip was considerable greater than that of the right so that the ratio of surface to volume was smaller on the left. Because of fluctuating brachial and digital blood pressures, those measured before warming the body were unsatisfactory.

After warming they were :—

	Brachial	4th digital	Gradient systolic	diastolic
Right	140 — 85	114 — 50	26	35
		5th digital 122 — 50	18	35
Left	138 — 85	4th digital 122 — 52	16	33
		5th digital 122 — 52	16	33

It is clear from the measurements, that the maximum bloodflow to the clubbed finger tip was moderately greater than that to the normal. Since surface area is the important factor in this determination, it is safe to disregard the result per unit volume. There was no significant change, however, in the digital arterial pressures, as between the clubbed and unclubbed fingers of the same hand or of different hands, and the gradients were all within normal limits. The increase in maximum blood flow in the clubbed finger must therefore be due to a decrease in the total vascular resistance, which implies increased calibre of the digital vessels.

Several points in this case deserve emphasis. In the few such cases (without recognisable clubbing) reported with autopsy (11, 17), there was dilatation of the arteries as well as the veins. Souques's (21) case of phlebectasia with unilateral clubbing includes no autopsy report. It is possible that in our case as well as in his, there was associated but unrecognised dilatation of the arteries as well as of the veins. In our case, the variations in the clubbing between the different fingers, the splinter hæmorrhage beneath one of the nails and the calcification seen in the soft tissues of the hand by X-ray are evidence in favour of this view.

Case 4.* W.C., a 48 year old man, was admitted to the hospital for the second time complaining of pain in the right upper chest, right shoulder and arm, and of cough. He had been well until 8 months previously, when he began to experience aching pain in the right upper chest anteriorly. Several weeks later, he noticed a spread of the pain to the right shoulder and arm with limitation of movement of the shoulder because of the pain. At his previous admission, bronchoscopy and X-ray revealed the presence of a carcinoma of the right upper lobe bronchus; a right pleural effusion was tapped and replaced by air. There was no clubbing at that time.

On examination, during his second admission, there was a right Horner's syndrome. There was dulness, diminished breath sounds and diminished fremitus over the right upper lobe and at the right base posteriorly. The right supra-clavicular fossa was found obliterated by a firm mass, fixed to the underlying structures. Movements of the right shoulder were limited. There was tenderness along the upper border of the right trapezius muscle and in the region of the second right rib anteriorly. The right external jugular vein was more distended than the left.

The right hand was slightly but definitely larger than the left and the patient was unable to flex the right fingers as completely as the left. The fingers of the right hand were measurably thicker throughout their length than those of the left and the tips of the fingers of the right hand were slightly but definitely clubbed, which was confirmed by the measurably greater diameters of the nails of the right hand. A roentgenogram showed no changes in the bones. There was hypæsthesia and hypalgesia of the dorsal surfaces of the 3rd and 4th distal phalanges of the right hand. Except for the sympathetic nervous system changes to be described, there were no other neurological abnormalities.

The fingers of the right hand were warmer and the veins on the dorsum of that hand more distended than on the left when the body was cool. When, however, the body was warmed by placing both feet in a water bath at 43 to 45°C. for ½ an hour, the left hand became warmer and the veins on the dorsum of the left hand more distended than on the right. These changes in temperature were repeatedly confirmed thermo-electrically as well as by measurements of heat elimination. The antecubital venous pressure measured by the direct method was 8·5 cm. of water on the left and 8·1 cm. on the right. When the body was thoroughly warmed in a heating cabinet, there was no sweating over the right side of the face and neck, or over the right upper extremity. The blood pressures before and after warming the body were as follows :—

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* Reported with the kind permission of Dr. Charles Miller.

	Before warming.			
	Brachial	Digital (2nd)	Gradient systolic	diastolic
Right	115 / 85	92 / 75	23	10
Left	110 / 85	92 / 80	18	5
	After warming.			
Right	105 / 80	85 / 75	20	5
Left	105 / 82	85 / 75	20	7

In this case, there was a pulmonary lesion which in itself is capable of producing bilateral clubbing. We have reason to believe that the increase in digital arterial pressure and decrease in gradient is present in bilateral clubbing of pulmonary origin before the clubbing becomes clinically manifest. In this case, there was such a decrease in gradient in both upper extremities, despite the fact that there was no clinically detectable clubbing in the fingers of the left hand.

It is known that nerve section (15) or neuritis (16) can accelerate clubbing in the fingers affected by the nerve lesion. In this case, it is likely that the pathological sympathectomy effected by the tumour, accelerated the clubbing on the right and that if the circulatory status remained unchanged for a sufficient length of time, clubbing of the other side would eventually have developed.

It is clear then, from these cases as well as from those reported by other workers (7, 14, 18), that there is no constant difference in bloodflow under ordinary environmental conditions between the normal and clubbed fingers in unilateral clubbing. There is also no constant increase in the maximum heat elimination and no change in the blood pressure gradient in these cases. There are three possible explanations for this difference between bilateral and unilateral clubbing. First, it is possible that the changes observed in the bilateral cases are due to chance and are not significant. The number of cases and the constancy of the results make this very unlikely. Second, it may be that the unilateral cases differ fundamentally in mechanism from the bilateral. Third, it is possible that the factors operating in bilateral and unilateral clubbing are initially the same, but that, in the unilateral cases, complicating factors subsequently obscure the original ones. Some of these complicating factors are arterial obstruction by thrombi, venous and lymphatic obstruction, and nerve lesions. Only further work, especially on the early course of these rare cases, can decide this issue.

Summary.

1. The response of the blood vessels in clubbed fingers to environmental temperature changes is qualitatively normal and the maximum heat eliminations of the hands of patients with clubbed fingers are within normal limits.

2. The maximum heat elimination and hence the bloodflow of the distal phalanges of clubbed fingers secondary to lung or congenital heart disease is usually increased.

3. The digital arterial pressure is increased and the brachial-digital arterial pressure gradient decreased in clubbing secondary to lung or congenital heart disease. In hereditary clubbing these pressures and gradients are normal.

4. The bloodflow of the unilaterally clubbed finger tip, as indicated by maximum heat elimination may be increased or decreased. No significant change was found in the blood pressure gradient except for a bilateral decrease in a case interpreted to be bilateral clubbing with acceleration of the process in the fingers of one hand due to a sympathetic nerve lesion.

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Fig. 2.
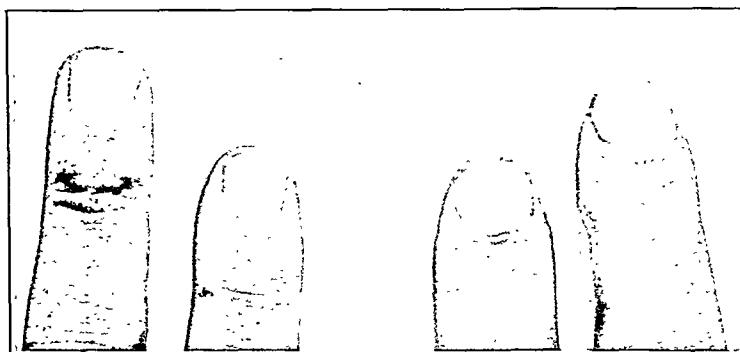
Fig. 3.
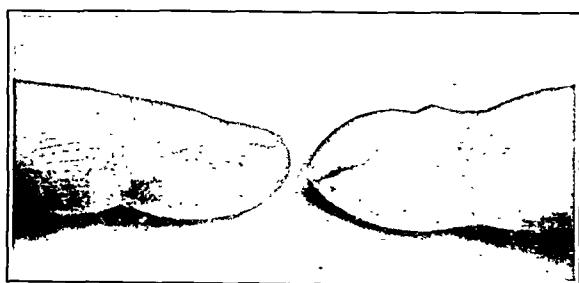
Fig. 4.

THE RATE OF BLOOD FLOW IN NORMAL FINGERS.*

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Introduction.

THE fingers have an extraordinarily rich vascular supply which is subject to marked vasomotor fluctuations. Studies of the rate of blood flow to the whole hand under various local and systemic conditions have been numerous and complete. It was desired, however, to determine the blood flow to the fingers without including the muscles of the hand. The method of Hewlett and Van Zwaluwenburg (4) has been adapted so that repeated quantitative measurements of the rate of blood flow to a single finger could be made every few seconds, care being taken to avoid reactive hyperæmia.

Method.

A Gærtner's capsule, namely, a finger blood pressure cuff, was placed around the proximal phalanx of the finger. A small, light, glass plethysmograph was then carefully fitted to the last two phalanges of the finger, as close to the cuff as possible, using thick vaseline to make the seal. Particular care was taken that there was no compression of the finger at the seal. This was verified repeatedly throughout the course of each experiment by showing that pressure within the cuff of as little as 10 mm. Hg. was sufficient to cause an increase in volume of the finger within the plethysmograph. The plethysmograph was connected by pressure tubing with a light tambour bearing a mirror which reflected the image of an illuminated slit on moving bromide paper. After the plethysmograph had been adjusted, the system was calibrated by injecting into it measured amounts (0·05 c.c.) of air from a graduated tuberculin syringe. It was found that the reflected beam moved in direct linear proportion to the amount of air injected, and that the pressure produced in the system never exceeded 5 cm. H$_2$O.. By means of a 3-way cock, air from a pressure tank at less than diastolic pressure could be suddenly

turned into or out of the Gærtner's capsule. The amount of this pressure was also recorded by means of a heavy tambour and mirror in the same optical system. Since the pressure was great enough to stop temporarily the venous outflow, but not great enough to impede arterial inflow, the finger swelled inside the plethysmograph and caused the reflected beam to move across the record. The rate at which it moved was a measure of the rate of blood flow into the finger. This could be calculated, using the volume of the finger in the plethysmograph, which was obtained at the end of each experiment by measuring the volume of water which the finger displaced. The calculated flow was expressed in c.c. per minute per 10 c.c. of finger volume. It was found that, using this method of measuring blood flow, results of great constancy could be obtained when the finger vessels were constricted and stable. With the vessels dilated and the flow great, the increase in volume of the finger following compression reached its maximum rapidly, within three to four pulse beats. In a large number of experiments it was found that the blood flow, though calculated from curves when only two or three pulse beats were available to measure from, gave remarkably constant results. It was also found in a study with fast recording that any distorsion produced by the compression occurred as the pressure in the cuff rose. Once the pressure in the cuff had reached its maximum, distorsion ceased (*see* Fig. 3) and consequently the first beat after the cessation of the act of compressing became available for calculation. The curves illustrated in Fig. 3 gave calculated values, in c.c. per minute per 10 c.c. finger volume, of 5·4 c.c., 4·3 c.c., 6·6 c.c., 5·4 c.c., 6·0 c.c.. Again, in four subjects in whom separate determinations were made on repeated occasions, as much as three months apart, there was close agreement in the rate of blood flow to the finger under similar conditions. It was, therefore, assumed that the method was one which permitted of considerable accuracy. A special water-jacketed plethysmograph was used for heating or cooling the finger locally. The finger itself was covered with water, but insulated from direct contact with the inner wall of the jacket by a perforated rubber mat. The temperature of the water in which the finger was bathed could be continuously taken by means of a thermojunction, and it could be maintained at any desired level by adjusting the flow and the temperature of the water in the jacket. (Fig. 1).

Plethysmographic measurements of the volume changes in an adjacent finger were made by the method of Bolton, Carmichael and Stürup (1). Skin temperatures were recorded from the finger pads by the use of small copper-constantan thermojunctions, constructed of gauge 28 wire. The galvanometer used permitted changes of 0·1°C. to be detected easily. Rectal temperatures were determined by a more sensitive galvanometer connected to a thermojunction placed 5 to 6 cm. above the anal sphincter.

Ten normal young adults served as subjects. The subject sat in a comfortable draught-free room maintained at a constant temperature (±1°C.). The hands were supported by arm rests at heart level. Body warming and vasodilatation in the hands were produced by immersing both

legs in water at 45°C.. Body cooling and vasoconstriction in the hands were produced by immersing the legs in water at 16° to 20°C.. After the body had been warmed and the vessels of the hands dilated, brief reflex vasoconstrictions were produced by stimuli previously shown to be effective,
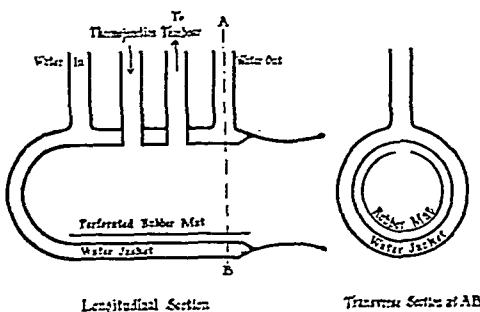

Fig. 1. Diagram of glass, water-jacketed finger plethysmograph for controlling local temperature. Water heated as required to maintain inner compartment at desired temperature is circulated through jacket. Temperature within inner compartment is taken by means of a thermo-junction led through a rubber cork at one of the middle openings, while the recording tambour is connected to the other opening. A perforated rubber mat separates finger from heated glass wall, but allows heat to pass freely to water bathing finger. For cooling finger locally, cold water may be circulated through jacket.

such as ice or pin-prick to an indifferent area of skin, a noise, the taking of a deep breath, or the solving of a mental problem (Stürup, Bolton, Williams and Carmichael (6)).

Results.

In establishing the validity of the blood flow method of Hewlett and Van Zwaluwenburg as here adapted to the finger two points of technique were early found to be necessary for success. It was first shown that there had to be no compression of the finger at the seal of the plethysmograph. This was especially important when the finger vessels were dilated and the flows very rapid. In this state three or four pulses were sufficient to distend the vessels to capacity, causing the curve subsequently to slope away from its upward direction towards the horizontal. If there was already congestion within the finger by compression at the seal, it was impossible to determine the true slope of the curve, owing to the limited capacity for further filling, so that only the later distorted portion of the curve was obtained. Secondly, it was necessary to place the cuff as close to the plethysmograph as possible without its moving the plethysmograph significantly when it was inflated. When the cuff was placed at a more distant site, as for example around the wrist, incorrect curves were obtained, due to the fact that the larger veins in the hand took up the blood from the finger which, therefore, did not swell in proportion to the flow to it. With attention to these two points consistent results were obtained.

Effect of body warming and body cooling. Many experiments were done on seven subjects to determine the rate of blood flow in the fingers when the cutaneous vessels were in a dilated and in a constricted state. In five of these experiments the blood flow was correlated with finger temperature. At the start of the experiment, when the subject felt quite comfortable and was neither warm nor cold, there were considerable spontaneous fluctuations in the rate of flow (Fig. 2). These variations in flow, especially when transient, were often unassociated with any change in finger temperature.
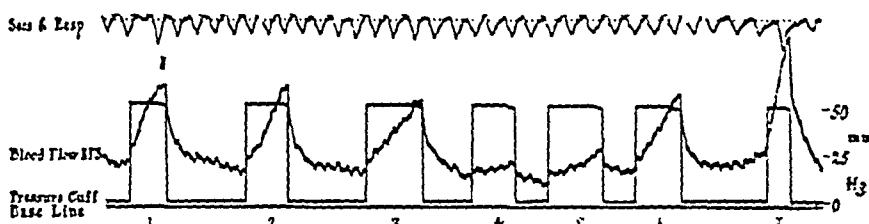
Fig. 2. Blood flow curves from right middle finger (Blood flow RF3). Subject comfortable, neither hot nor cold. In respiratory tracing (Resp.) down stroke represents inspiration. Pressure within congesting cuff (Pressure cuff) is in mm. Hg., as shown on appended scale. These are the same for all records.

Calculated values in c.c. per minute per 10 c.c. finger volume :—Curve No. 1, 1·0 c.c.; No. 2, 0·9 c.c.; No. 3, 0·5 c.c.; No. 4, 0·08 c.c.; No. 5, 0·18 c.c.; No. 6, 0·66 c.c.; No. 7, 2·0 c.c.
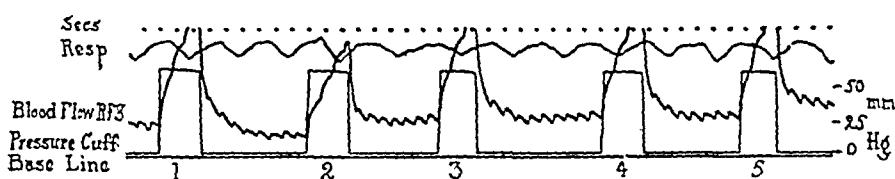

Fig. 3. Blood flow curves from right middle finger (Blood flow RF3). Subject warm and vessels dilated. Same experiment as in Fig. 2. Calculated values in c.c. per minute per 10 c.c. finger volume :—Curve No. 1, 5·4 c.c.; No. 2, 4·3 c.c.; No. 3, 6·6 c.c.; No. 4, 5·4 c.c.; No. 5, 6·0 c.c.

Fig. 3 shows a typical series of curves recorded after the subject's legs had been immersed in hot water. This procedure had warmed the body and produced vasodilatation in the fingers, so that the finger temperature remained high and steady. The rate of blood flow to the finger was then found to be greatly increased and much more constant than before, so long as a vasoconstricting stimulus was not given. Fig. 4 shows a series of curves obtained after immersion of the legs in cold water, which caused the vessels to become constricted and the finger temperature to fall. The rate of flow in this state was very low but again quite constant.

Fig. 5 gives the time relations of the events in the experiment in which the above records were obtained. Each measurement of blood flow is an average of at least three separate determinations. During the control

period, when the rectal temperature was constant, there was considerable fluctuation in the blood flow and in the temperature of the finger. With the rise of rectal temperature after putting the legs in hot water, vasodilatation in the fingers occurred as shown by the marked increase in finger blood flow and finger temperature. When the legs were then immersed in cold water there was a sharp vasoconstriction as measured by the blood flow, then a temporary relaxation, followed by a lasting and more profound
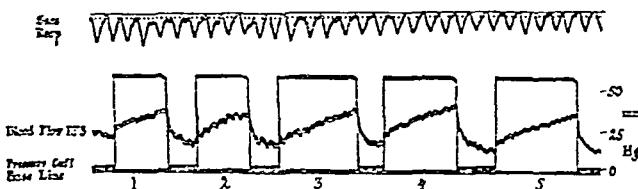
Fig. 4. Blood flow curves from right middle finger (Blood flow RF3). Subject cold and vessels constricted. Same experiment as in Fig. 3.
   Calculated values in c.c. per minute per 10 c.c. finger volume :—Curve No. 1, 0·13 c.c.; No. 2, 0·20 c.c.; No. 3, 0·16 c.c.; No. 4, 0·15 c.c.; No. 5, 0·13 c.c.
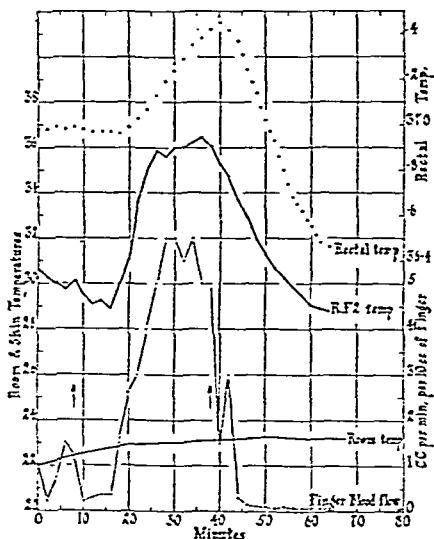

Fig. 5. Blood flow to right middle finger (Finger blood flow), temperature of tip of right forefinger (RF2 temp.) and rectal temperature (Rectal Temp.) during body heating and cooling. At 8 minutes (first arrow) both legs were immersed in water at 45°C.. At 38 minutes (second arrow) the legs were immersed in water at 20°C..

constriction. The finger temperature showed a smooth and more gradual fall.

Table I gives the average rates of blood flow to the fingers with the vessels constricted and dilated in the seven subjects on whom these experi-

ments were carried out. The rate of blood flow to the finger in norm: subjects ranged from 0·02 c.c. per minute per 10 c.c. when the periph . vessels were constricted, to 12 c.c. per minute per 10 c.c. of finger, a dilated. In four subjects in whom separate determinations were made c repeated occasions, as much as three months apart, there was close agreemen in the rate of blood flow to the fingers under similar conditions. In fiv subjects in whom simultaneous determinations of the flow to correspondin; fingers of each hand were made, there was practically an identical flo, on the two sides. When any significant difference did exist in the prelimin. stage of the experiment it usually disappeared after the two hands had be.. brought to the same state of full vasodilatation by body warming. Howeve in two subjects there persisted a slightly greater flow in the fingers of the right hand than of the left.

TABLE I.

Finger blood flow in c.c. per min. per 10 c.c. finger volume.

Subject.	Constricted.	Dilated.
1	0·10 c.c.	9·0 c.c.
2	0·45 c.c.	8·0 c.c.
3	0·15 c.c.	10·0 c.c.
4	0·15 c.c.	7·8 c.c.
5	0·02 c.c.	6·0 c.c.
6	0·07 c.c.	12·0 c.c.
7	0·70 c.c.	6·0 c.c.

Effect of transient vasoconstrictions. It is known that when the vessels of the digits are already dilated as a result of body warming certain stimuli, such as pin-prick, or ice to the skin, noise, a deep inspiration, or the solving of a mental problem, cause sharp vasoconstrictions which are mediated by impulses in sympathetic nerves (Stürup, Bolton, Williams and Carmichael (6)). So brief are these vasoconstrictions that they cause barely discernible changes in skin temperature. Blood flow determinations, however, have demonstrated that marked decreases in blood flow occur before and after a stimulus. It was found that at about the third second after the stimulus flow was diminished, was slowest at about the sixth second, and at about the tenth second began to return to the control level. The calculated values were as follows :—before the stimulus 8·0 c.c., at the third second 4·6 c.c., at the sixth second 2·1 c.c., and at the tenth second 6·0 c.c.. The decrease in blood flow to the finger following the stimulus was simultaneous with a decrease in the volume of an adjacent finger. In over 200 estimations of

this effect on ten subjects it was found that such vasoconstricting stimuli regularly caused temporary reductions in the blood flow to the fingers, at times to as much as one-twentieth of the previous rate.

Effect of local heat. By means of the special water-jacketted plethysmograph (Fig. 1) it was possible to heat a single finger locally without altering the body temperature or the rate of flow to the remaining fingers. In this way the dilating effect of local heat alone, without any possibility of sympathetic reflex activity, could be studied. It was first demonstrated that under normal conditions the flow to the corresponding fingers of the two hands was the same, both with the finger vessels constricted and dilated. It was also shown that when the finger vessels were already dilated, a constricting stimulus (deep breath) reduced the flow equally on the two sides. The body was then cooled by immersing the legs in cold water and after the flows had become greatly reduced, one finger alone was heated to 45°C.. The flow in this finger became markedly increased, but the flow in the control finger remained the same. The flow in the heated finger, however, was not so great as it had been when the body was warmed, and it increased farther when the body was again warmed by immersing the legs in hot water. There was then little difference between the rate of flow on the heated and on the normal sides, but when a vasoconstricting stimulus was given the reduction in blood flow was much less in the heated than in the normal finger.

Flow in different phalanges. Since with the method used blood flow to the two terminal phalanges only was measured, the results obtained cannot be taken as quantitatively correct for the whole finger. Indeed, it was possible to show that in the dilated state the rate of flow to the terminal phalanx was much greater than to the middle phalanx. The blood flows in two corresponding fingers were first shown to be the same. The plethysmograph was then adjusted on one finger so as to include only the terminal phalanx. It was found, in spite of the fact that the volume of finger in the plethysmograph was halved, that the flow was diminished by only one quarter. The circulation to the terminal phalanx was then occluded by a tight elastic band, and the plethysmograph re-adjusted to include the middle phalanx. The flow was now diminished by three-quarters. Calculated on a basis of volume, the flow to the middle phalanx was about one-third of the flow to the terminal phalanx. These proportions varied from subject to subject, but the terminal phalanx always had a greater flow. When the flow in the terminal phalanx or in the middle phalanx was being determined separately, it varied in the same direction in both in response to vasodilating and vasoconstricting influences.

Discussion.

The present method of determining the blood flow in the finger has allowed a quantitative measurement of the wide fluctuations which occur physiologically in response to vasodilating and vasoconstricting influences.

It has been shown that blood flow to the finger may increase 100 times (for example, from 0·1 c.c. to 10 c.c. per minute per 10 c.c. of finger volume) in a few minutes during body warming, and may temporarily decrease 20 times in the few seconds following a vasoconstricting stimulus. Not only because it allows quantitative measurements, but also because it allows measurement of considerable accuracy of rapid and transient changes, this method has been found superior to the usual skin temperature method which ` has an appreciable lag, and is limited by the differential between blood and room temperature.

It is of interest that the dilatation in the finger vessels resulting from body warming alone is more complete than that from local heating of the finger alone. The dilatation in the fingers produced by body warming alone was almost as great as that produced by body warming together with local heating of the finger. It should also be noted that local heat is effective in inhibiting but not in completely abolishing the vasoconstrictions in the finger caused by stimuli when the vessels are already dilated by body warming.

The observation that blood flow to the terminal phalanx of the finger is considerably greater than to the second phalanx is consistent with the findings of Lewis (5) and of Grant and Bland (2), who found that the finger tip and nail bed warmed more quickly and more markedly under vasodilating influences than the middle phalanx. Grant and Bland (2) related this difference to the anatomical distribution of the arteriovenous anastomoses in the finger, which they found to be most numerous in the terminal phalanx. While it is found that the blood flow is quantitatively greater in the terminal phalanx than in the middle phalanx, no qualitative difference is found in the alterations in the flow to the two parts in response to vasodilating or vasoconstricting influences. Grant and Pearson (3) have pointed out that with the peripheral vasodilatation caused by body warming the blood flow to the finger is greater than the flow to the hand and to the hand greater than to the forearm. The present study has extended this work to the separate parts of the finger, when again a greater flow was found to the distal than to the proximal parts. The discrepancy in the actual values for blood flow to the finger after vasodilatation reported by Grant and Pearson (2 to 3 c.c. per minute per 10 c.c. of finger) and the values here reported (5 to 12 c.c. per minute per 10 c.c. of finger) may be partly due to the fact that Grant and Pearson measured the whole finger and not merely the two terminal phalanges. It is also possible that the discrepancy may in part be due to the fact that they placed the congesting cuff at the wrist.

The difference in blood flow to the different parts of the finger must be recognised in using the method to estimate the flow to the fingers in a given case. By measuring as a routine the flow to both terminal phalanges, however, a normal range has been established with which pathological cases may be compared. The wide normal range from the very slow flow when the vessels are constricted to the very rapid flow when they are dilated makes

obvious the necessity for accurately controlling the conditions under which the measurements are made. When the conditions are definitely the same on two occasions (for example, either full dilatation or full constriction) very similar results may be obtained. With such precautions, the method may prove useful in quantitatively measuring from time to time and in various stages of dilatation and constriction the blood flow to fingers affected with vascular or vasomotor disturbances.

SUMMARY.

The method of Hewlett and Van Zwaluwenburg has been adapted to estimate blood flow in the finger.

It has been shown that the blood flow to the finger may be increased as much as 100 times during the vasodilatation produced by body warming.

Following a vasoconstricting stimulus when the vessels are already dilated, the flow to the finger may be temporarily decreased as much as 20 times.

The increase in blood flow resulting from local heating of the finger is not so great as that produced by body warming.

The blood flow to the terminal phalanx is considerably greater than to the middle phalanx of the finger when the vessels are dilated.

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THE INHIBITION OF PITUITARY ACTIVITY IN ACROMEGALY BY OESTRADIOL BENZOATE AND TESTOSTERONE PROPIONATE.

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Two procedures have been available in the past for reducing the activity of the pituitary gland in acromegaly, namely excision of the eosinophile tumour, and deep X-ray irradiation. Increasing knowledge of the relations of the endocrine glands has suggested that pituitary activity may be modified by the injection of the sex hormones. After gonadectomy the anterior pituitary lobe hypertrophies and its function increases. Marrian and Butler (5) showed that œstrogens decrease pituitary activity in large doses and stimulate activity in small doses. Kirklin and Wilder (4) used this principle in an attempt to treat cases of acromegaly. They injected their cases with small doses of œstrogens, and judged their results by clinical observation. A more objective method would be preferable. Schrire and Zwarenstein (8 and 9) showed, in animals, that pituitary activity may be judged by the quantities of creatine and creatinine in the urine. Schrire (6) investigated cases of acromegaly and found that creatine and creatinine excretion was abnormally increased, and that the altered metabolism of these substances appeared to be characteristic of the disease. Schrire and Zwarenstein (10 and 11) injected testicular and ovarian extracts into rabbits, and were able to demonstrate alterations in creatinine excretion which were attributed to pituitary inhibition. It was decided, therefore, to treat patients with acromegaly by injections of œstradiol benzoate and testosterone propionate.

Methods.

Quantitative estimations of creatine and creatinine in the urine were determined by Folin's colorimetric method (2). The measures adopted for the collection of the urine have been detailed in a previous communication (6). The diet throughout the period of investigation of each patient was

prepared by a dietitian and was creatine free. Oestrogenic substance was administered as œstradiol benzoate (Organon), 1 c.c. of which contains 5 mg. (50,000 international units). The dose was 10 mgms. intramuscularly
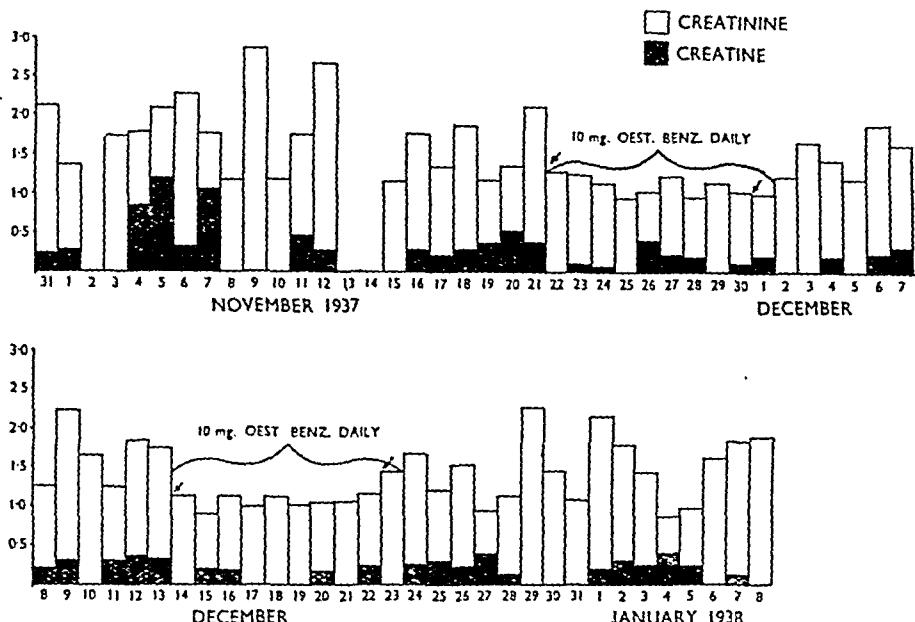

Fig. 1. Case 1. Acromegaly. Chart of urinary creatine and creatinine output in g. per 24 hrs.
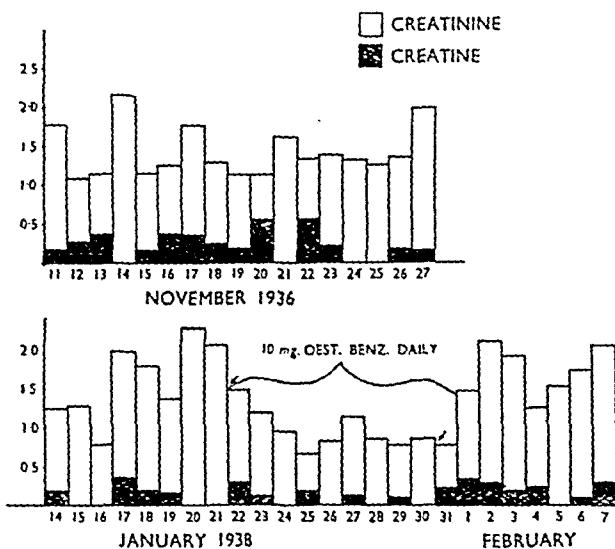
Fig. 2. Case 2. Acromegaly. Urinary creatine and creatinine output in g. per 24 hrs.

each day. Testosterone propionate (Ciba), 1 c.c. of which contains 50 mg., was injected intramuscularly in doses of 100 mg. a day. The dosage of both substances was chosen arbitrarily. Four patients with acromegaly,

two male and two female, have been investigated; the case reports are appended.
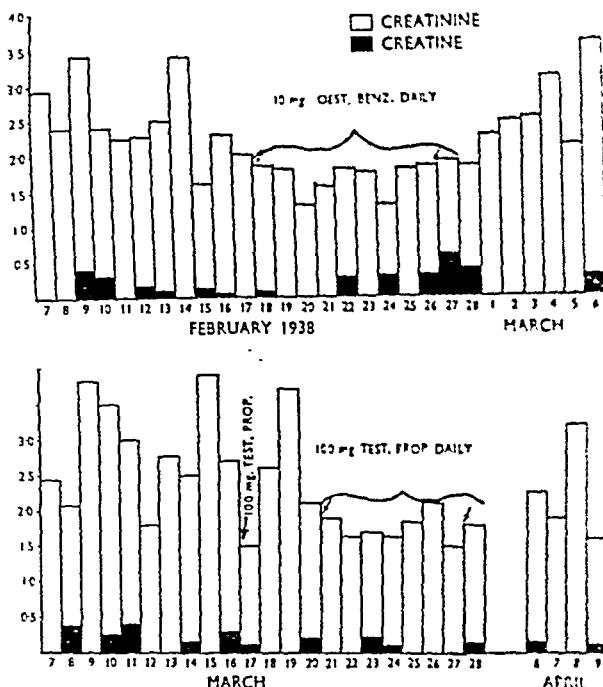
Fig. 3. Case 3. Gigantism and acromegaly. Urinary creatine and creatinine output in g. per 24 hrs.
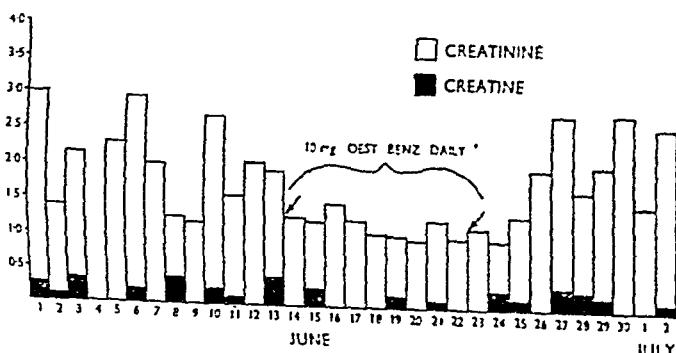

Fig. 4. Case 4. Acromegaly. Urinary creatine and creatinine output in g. per 24 hrs.

Results.

The 24 hour excretion of creatine and creatinine in the urine was estimated in each case, and the results are given in the charts. During a preliminary control period all four patients showed an excessive excretion

of creatinine and an irregularity in the amount excreted. Following the control period, each patient was injected with 10 mg. of œstradiol benzoate daily for 10 days. In each case, within 24 or 48 hours of starting the injections, the excretion of creatinine fell to a lower and approximately normal level and during the period of injection the daily excretion ceased to fluctuate. Within 1 to 3 days of stopping the injections the creatinine excretion again became excessive and varied in amount from day to day. In Case 1 the ten day period of injection was repeated, and a similar reduction in the excretion of creatinine was observed.

In Case 3 100 mg. of testosterone propionate were injected daily for 8 days, a trial dose having been given four days previously. The injections were started 23 days after stopping the œstradiol benzoate. For these 23 days he showed the excessive, irregular creatinine output that had been observed during the control period. However, for the 8 days that testosterone propionate was given, creatinine excretion was at a constant level and the amount excreted daily was comparable with that noted during the period of œstradiol injection. The patient had then unavoidably to leave hospital; 10 days later, when collection of urine was again possible the excretion of creatinine appeared, once more, to be excessive and irregular.

The excretion of creatine in these patients during the control periods was in excess of that found in normal individuals. The injections seemed to have no significant effect on creatine excretion.

Clinical symptoms during the periods of injection were noted in one case only. Case 3 complained of increase in the frequency and severity of his headaches during the injection of both œstradiol and testosterone. It is possible that in this patient some change in size of the pituitary gland may have occurred and have been sufficient to produce symptoms. He also noticed an increase of sexual desire with œstradiol and had two nocturnal emissions during this period. Testosterone produced no increase of desire, but towards the end of the period of injections there was a tendency to spontaneous erections never amounting to priapism.

Discussion.

The daily excretion of creatinine in normal individuals, on a creatine free diet, is relatively constant (3). The amount of creatinine excreted is independent of the urinary volume but is related to the weight and muscular build of the individual. The irregular, excessive creatinine output of the control periods in these four cases of acromegaly confirm the results previously obtained in nine other patients (6). The injection of œstradiol benzoate, and in one case testosterone propionate, into these four patients appeared in every instance to cause creatinine to be excreted in smaller and more constant amounts, amounts compatible with the excretion of a normal individual of the same weight. A rapid return to the original excessive and irregular excretion happened when the injections were stopped. Schrire and Sharpey-Schafer (7) have recently published observations which suggest that the

excessive creatinine excretion in acromegaly is associated with the gonadotropic hormone of the pituitary gland ; normal individuals injected with a preparation of pituitary gonadotropic hormone showed a rise in the excretion of creatinine. No rise was produced by pituitary thyreotropic and growth principles or by the gonadotropic substances that are prepared from pregnancy urine and mare's serum in the doses employed. It would appear, therefore, that the injection of œstradiol or testosterone into cases of acromegaly suppresses an abnormality of metabolism that is associated with the gonadotropic hormone of the overactive pituitary gland. This conclusion would be in keeping with the findings of Marrian and Butler and of Schrire and Zwarenstein in animals. The change in creatinine excretion produced by œstradiol can be correlated with the observation that the creatinine output of a case of acromegaly fell from an excessive irregular level to a constant lower level after excising the eosinophile tumour (6).

It has been suggested (7) that the excretion of creatine in excessive quantity by patients with acromegaly may be due to an overproduction of thyreotropic hormone stimulating the thyroid gland. In these four cases no significant change in creatine excretion was found to result from the injections.

SUMMARY.

1. The increased elimination and fluctuating output of urinary creatine and creatinine in acromegaly has been confirmed.

2. The effect of injecting large doses of the sex hormones in four cases of acromegaly has been studied.

3. Large daily doses of œstradiol benzoate reduced the creatinine in the urine to normal levels. During the period of injection the creatinine excretion ceased to fluctuate and was constant as in the normal.

4. Large doses of testosterone in one case produced similar results.

5. It is suggested that an abnormality in acromegaly associated with gonadotropic pituitary activity can be suppressed by the administration of the sex hormones.

CASE REPORTS.

Case 1. E.M.R. Female, aged 34. Weight 140 lbs. Height 5ft. 2ins. In 1936 her hands and feet began to enlarge and in the next 5 years she gained 40 lbs. in weight. In 1930 coarse hair began to grow on her face, trunk, arms and thighs. Her voice gradually altered, becoming hoarse and deep, the breasts became smaller but menstruation was regular. She complained of periodic headache. In 1933 the thyroid gland was found to be enlarged and a nodular goitre was removed in July, 1937. The daily urinary output was small and the presence of an anti-diuretic substance was demonstrated in the cerebro-spinal-fluid (1).

In October, 1937, she was admitted to Hammersmith Hospital. Her appearance was typical of acromegaly. The skin of the face was coarse and thickened with some degree of acne. The nose was large, prognathism was marked, and the teeth were widely spaced. The hands and feet were large, and the fingers shortened and thickened. There was some kyphosis, but no enlargement of the viscera could be detected clinically. The visual fields and fundi were normal. X-rays showed ballooning of the sella turcica with thinning of the floor and clinoid processes, and tufting of the terminal phalanges. She was very hirsute, the hair being distributed as in the male. The breasts were small and the general aspect of the patient conformed to that of the

male. No abnormality of the internal and external genitalia was found. X-rays showed no opaque deposits in the suprarenals. Menstruation had not occurred since the thyroidectomy 4 months previously. Headaches were frequent, being bilateral and frontal in type. The basal metabolic rate varied between + 26% and + 12 %. The urine, blood count, and the glucose tolerance test were normal. The chemical constituents of the blood (urea, chloride, calcium, plasma protein and phosphatase) were within normal limits. The blood cholesterol was elevated to 370 mg. per 100 c.c.. The mental state of the patient was peculiar and she showed definite psychotic tendencies associated with systematised delusions.

Case 2. L.N. Female of 29 years. Weight 160 lbs. Admitted to the National Hospital, Queen Square, under the care of Dr. E. Arnold Carmichael.

In 1927 she began to be troubled with headaches and loss of vision. At that time she noticed an increase in the size of her hands and feet, and prognathism was present. The vision was restricted in both temporal fields. In the same year, Radon seeds were implanted in the pituitary gland. In 1929, deep X-rays to the sella turcica were given, and 6 months later she was operated on and part of the pituitary tumour was removed. She was readmitted in 1936 for investigation of her creatine and creatinine excretion, and once again in 1938 for the present investigation.

The skin of the face was thick and coarse. Hirsutism was marked. Prognathism was obvious and the teeth of the lower jaw were widely spaced. The hands were thick and spade shaped, and the feet were large. Menstruation was regular, but the flow was scanty. The urine and the glucose tolerance test were normal. X-rays showed an enlarged pituitary fossa and erosion of the clinoid processes.

Case 3. H.A.W. A man of 29 years. Weight 222 lbs. Height 6ft. 6½ins. His father is 6ft., and his two brothers are 6ft. 1in., and 6ft. 3ins., respectively. At 17 years of age he was 5ft. 11ins.. In 1929 at the age of 20 he began to grow quickly and reached his present height of 6ft. 6½ins at the age of 26. In 1933 he first noticed his hands were growing larger and his nose began to increase in size. At this time bitemporal headaches began. The diagnosis of acromegaly was made in 1936. He received deep X-ray therapy to the pituitary gland. For 6 months there was considerable improvement, and the frequency and severity of the headaches abated. More recently the headaches have become prominent again. Puberty was delayed until he was 17 years old. Nocturnal emissions were rare.

On admission in February, 1938, the facies were typical of acromegaly. The nose was large, the lips thick and fleshy and the teeth well spaced. The hands were large and the fingers long and broadened. The feet were very big. No changes in vision had been noticed, and the visual fields and fundi were normal. The glucose tolerance test was normal, the B.M.R. — 23%, the E.C.G. normal and the urine showed nothing abnormal. X-rays showed a normal sella turcica. The epiphyses of the limbs were united.

Case 4. L.L. A man of 72 years. Weight 134 lbs. Height 5ft. 3¼ins.

For 30 years he had increasing loss of vision. Twelve years ago bilateral iridectomy was performed for chronic glaucoma and later the left eye was removed. He complained of headaches for 15 years, but was not aware of any change in his appearance. Admitted in June, 1938, he presented characteristic features of acromegaly. The nose and lips were large and coarse, the tongue enlarged and prognathism was present. The fingers were short, broad and thick, and the hands wide. Similar changes were present in the feet. There was some kyphosis. The left eye was absent, the right eye showed evidence of an old iridectomy, the fundus could not be clearly seen, and vision was poor especially in the nasal field. The small muscles of the left thenar eminence were wasted without any sensory loss or other abnormal sign in the nervous system. X-ray showed a large sella turcica and large frontal sinuses. The hands showed tufting of the terminal phalanges. Blood chemistry was normal. The urine and the glucose tolerance test were normal.

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